# entomon

#### A Quarterly Journal of Entomological Research

Vol. 12

MARCH 1987

No. I



PUBLISHED BY
THE ASSOCIATION FOR ADVANCEMENT OF ENTOMOLOGY
DEPARTMENT OF ZOOLOGY, UNIVERSITY OF KERALA, KARIAVATTOM
TRIVANDRUM, INDIA 695 581

#### **ENTOMON**

Entomon is a quarterly journal of the Association for Advancement of Entomology issued in March, June, September and December, devoted to publication of research work on various aspects of insects and other land arthropods.

#### EDITORIAL ADVISORY BOARD

T. N. ANANTHAKRISHNAN, Institute of Entomology, Madras (Chairman)
M. D. PATHAK, International Rice Research Institute, Philippines
K. N. SAXENA, International Centre for Insect Physiology and Ecology, Nairobi

#### EDITORIAL BOARD

K. J. JOSEPH, University of Calicut, Calicut

V. S. KAVADIA, Sukhadia University, Jaipur

G K. Manna, University of Kalyani, Kalyani

K. N. MEHROTRA, Indian Agricultural Research Institute, New Delhi

N. MOHANDAS, Kerala Agricultural University, Trivandrum

M. R. G. K. NAIR, Kerala Agricultural University, Trivandrum

M. K. K. PILLAI, University of Delhi, Delhi

N. R. PRABHOO, University of Kerala, Trivandrum

V. K. K. PRABHU, University of Kerala, Trivandrum (Managing Editor)

P. S. RAMAMURTY, University of Hyderabad, Hyderabad

Address MS and all editorial correspondence to Managing Editor, *Entomon*, Department of Zoology, University of Kerala, Kariavattom, Trivandrum, India 695 581.

#### SUBSCRIPTION RATES

Annual	Subscription	for	institutions:	Rs 150/- (	in India)	); <b>{</b>	50/-	(al	broad,	air	mail)
,,,	,,		individuals:	Rs 50/-	,,	; {	20/-	(	"	sea	mail)

© 1987 by the Association for Advancement of Entomology. All rights reserved.

- 1. All remittance to the journal or the Association should be sent to the Secretary-Treasurer of the Association by bank draft only, A/c payee in favour of the Association for Advancement of Entomology.
- 2. Requests for replacement copies of ENTOMON in lieu of numbers lost in transit, should reach the Secretary-Treasurer not later than three months after the date of publication of the number.

### entomon

Vol.	12	March	1987	Number	1

CONTENTS

CONTENTS
Host-specificity of Cyrtobagous salviniae Calder & Sands (Col., Curculionidae) introduced into India for the control of Salvinia molesta—K. P. Jayanth and Sudha Nagarkatti
Protein and glycogen contents of the accessory reproductive glands of the male and female silk moths Bombyx mori before and after mating—  S. H. Bhosale, V. L. Kallapur and K. Venkatesh
Effect of Catharanthus alkaloids on the house fly, Musca domestica L — Kumuda Sukumar
Effectivenss of spray formulations against rice leaf folder Cnaphalocrocis medinalis Guenee (Lepidoptera: Pyralidae)—Mangal Sain, N. V. Krishnaiah and M. B. Kalode
Day biting mosquitoes (Diptera: Culicidae) of Manipur—K. B. Rajput and T. K. Singh
Growth pattern of Corcyra cephalonica pupa and effect of diflubenzuron on pupal weight—M. Sriramulu and K. N. Mehrotra
The metabolism and excretion of C-labelled dieldrin in resistant and susceptible <i>Tribolium castaneum</i> (Herbst)—A. S. Udeaan and R. L. Kalra
Howmany larval instars are there in Opisina arenosella?—P. B. Santhosh Babu and V. K. K. Prabhu
Observations on spiders (Order: Araneae) predacious on the coconut leaf eating caterpillar Opisina arenosella Wlk. (Nephantis serinopa Myrick) in Kerala: Feeding potential—B. Sathiamma, S. P. Jayapal and G. B. Pillai
A new genus and three new species of eriophyid mites (Acarina: Eriophyoidea) from West Bengal India—N. K. Ghosh and S. Chakrabarti
Effect of mating duration on fecundity and fertility of eggs in Bombyn mori L. (Lepidoptera: Bombycidae—M. Thomas Punitham, M.A. Haniffa and S. Arunachalam
Studies on host age preference and biology of exotic parasite, Cotesia marginiventris (Cresson) (Hymenoptera: Braconidae)—S. K. Jalali, S. P. Singh and Chandish R. Ballal
Studies on the host selection and discrimination behaviour of Diaeretiella rapae (M'intosh), a parasitoid of Lipaphis erysimi (Kalt).—S. C. Dhiman and Vinay Kumar.

#### **BRIEF COMMUNICATION**

A note on the occurrence of Anopheles minimus Theobald in Manipur—	
K. B. Rajput and T. K. Singh	43
In memorium—K. J. Joseph	69
BOOK REVIEW	74
ANNOUNCEMENTS	

#### **Author Index**

Arunachalam, S., 55	Mehrotra, K. N., 29
Ballal, R., 59	Nagarkatti, S., 1
Bhosale, S. H., 7	Pillai, G. B., 45
Chakrabarti, S., 49	Prabhu, V. K. K., 39
Dhiman, S. C., 67	Punitham, M. T., 55
Ghosh, N. K., 49	Rajput, K. B., 21, 43
Haniffa, M. A., 55	Sain, M. 17
Jalali, S. K., 59	Santhosh-Babu, P. B., 39
Jayanth, K. P., 1	Sathiamma, B., 45
Jayapal, S. P., 45	Singh, S. P., 59
Joseph, K. J., 69	Singh, T. K., 21, 43
Kallapur, V. L, 7	Sriramulu, M., 29
Kalode, M. B., 17	Sukumar, K., 13
Kalra, R. L., 13	Udeaan, A. S., 33
Krishnaiah, N. V., 17	Venkatesh, K., 7
Kumar, V., 67	

#### ACKNOWLEDGEMENT OF GRANT

Association for Advancement of Entomology is grateful to the Department of Science and Technology, New Delhi, for a grant of Rs. 15,000/- for the year 1986—1987 for publication of ENTOMON.

## HOST-SPECIFICITY OF CYRTOBAGOUS SALVINIAE CALDER & SANDS (COL., CURCULIONIDAE) INTRODUCED INTO INDIA FOR THE CONTROL OF SALVINIA MOLESTA

#### K. P. JAYANTH & SUDHA NAGARKATTI\*

All India Coordinated Research Project on Biological Control of Crop Pests and Weeds, Indian Institute of Horticultural Research, Bangalore, India 560 089

(Received 7 August 1985;

The host-specificity of Cyrtobagous salviniae Calder & Sands (Col. Curculionidae), imported for biological control of the aquatic weed Salvinia molesta in India, was studied. Seventy five species of plants representing 41 families were tested. The weevil did not feed or reproduce on 67 of the test plants. Extremely limited feeding was observed on Amorphophallus sp., Colocasia esculenta, Ipomaea batatas, Lactuca sativa and Raphanus sativus, with slightly more on Pistia stratiotes and Trapa bispinosa; but damage was negligible and no oviposition occurred on any of these plants. In the presence of Salvinia, none of the above plants were attacked, clearly showing that C. salviniae safe for introduction. Following these studies, C. salviniae was introduced into a lily pond at Bangalore and field releases were also initiated in Kerala. The weevil is now established as evidenced by the presence of all immature stages and freshly emerged adults. (Key words: Cyrtobagous salviniae, Salvinia molesta, Biological control)

#### INTRODUCTION

The free-floating aquatic plant Salvinia molesta Mitchell is native to Brazil (FORNO & HARLEY, 1979) and is believed to have spread to Asia and Africa through introduction into botanical gardens. Salvinia was first observed in Kerala in 1955 and since 1964 has assumed pest status (Joy, 1978). According to Joy the weed is now present all over Kerala and in the Kuttanad district alone some 75,000 acres of canals, rivers, ponds and other water bodies are infested. Navigation and irrigation are adversely affected, besides fishing, and other operations requiring unimpeded flow of water.

In some areas cultivation of paddy is abandoned on account of *Salvinia* infestation. Efforts to introduce *Paulinia* acuminata DeGeer in Kerala have not been successful, the grasshopper being subject to predation by frogs, toads, spiders etc. (Joy et al., 1981), although it was considered a potential control agent by FORNO (1981).

Cyrtobagous salviniae CALDER & SANDS (earlier believed to be singularis Hustache), a weevil belonging to the aquatic tribe Bagoini, was one of three insects recommended by BENNETT (1966). Introduction of this insect originating from Southeastern Brazil into Lake Moondarra covering 400 ha in northern Queensland (Australia) in June 1980, resulted in complete control of the weed within 18 months (Room et al., 1981). SANDS (1983)

Contribution No. 101/85 of the Indian Institute of Horticultural Research, Bangalore.

<sup>\*</sup> Present address: 1113 J Blue Heron Drive, Pineville, N. C. 26134, U. S. A.

reported that the weevil population originating from Brazil differs from C. singularis collected from Trinidad by BENNETT (1966) and was later identified as C. salviniae (CALDER & SANDS, 1985). A stock of this weevil was received from Australia through the courtesy of Dr. K. L. S. HARLEY of Long Pocket Laboratories, Indooroopilly, Australia. The insect culture was established in quarantine at Bangalore and host-specificity tests were carried out to reconfirm the tests that had earlier been carried out by the Indian Station of the Commonwealth Institute of Biological Control, Bangalore (SANKARAN & RAMASESHAIAH, 1973).

#### MATERIALS AND METHODS

The shipment of C. salviniae was received in July 1982. Ten adults were released on yong plants of Salvinia held in 20 × 16 cm clear plastic jars half filled with water and with lids suitably modified to provide aeration. After 5-7 days, the adults were collected back and released on fresh plants and the process repeated until they died. The exposed plants were held in 61×40.5×30.5 cm opaque plastic troughs filled with water collected from an outdoor tank. The troughs were kept covered with cloth cages provided with aeration holes covered with nylon mesh sleeves for easy handling and polythene sheeting at the top to admit light, the whole being supported on a wrought iron frame.

Seventy-five species of plants representing 41 families were each exposed to freshly emerged adults of C. salviniae sp. for host-specificity tests. For the no-choice tests (in which individual plants were tested in the absence of the natural host), bouquets of the test plant were placed in plastic jars, with the stems dipping in water contained in small vial and with a cotton wick projecting from the vial to provide free water for the adults to feed on. Aquatic test plant were held directly in water. Five adults were released per plant and observations were recorded until they died. Yellowing or decaying plants were replaced whenever required.

Observations were recorded on survival, feeding and oviposition. In the multiple-choice tests, all plants on which some nibbling or feeding had been observed were provided together with a single Salvinia plant. The tests were replicated three times.

#### RESULTS AND DISCUSSION

Under laboratory conditions, *C. salviniae* completed development in about 45 days. Adult weevils survived upto 60 days, feeding on tender leaves and making circular feeding holes. Newly emerged adults were unable to survive in the absence of fresh growth of *Salvinia*. Eggs were laid within the plant tissues or suspended in the root mass. The larvae mined through the stolons and leaf axils causing rapid decay and degeneration of the plants.

C. salviniae did not feed or oviposit on any of the 67 plants listed in Table 1, even though they survived for 6 to 30 days. Slight nibbling was observed on 8 plants (Table 2). One feeding spot was observed on Amorphophallus sp., and 2-5 spots on Colocasia esculenta, Ipomaea batatas, Lactuca sativa and Raphanus sativus. Among the aquatic plants, 3 feeding spots were observed on Nymphaea sp. There were more feeding spots on Pistia stratiotes and Trapa bispinosa, but damage to the plants was negligible. Also, oviposition did not occur on any of the above plants.

In the multiple choice tests feeding was observed only on *Salvinia*, indicating that in the presence of the natural host, none of the test plants on which nibbling was observed in a no-choice situation would be attacked. Earlier studies at the CIBC West Indian Station (BENNETT, 1966) and Indian Station (SANKARAN & RAMASESHAIAH, 1973) had also indicated that the weevils would not feed on or

TABLE 1. Test plants on which feeding by C. salviniae was not observed.

S. No. Family		Species	Common name	Max. No. of days survived
1	Amaryllidaceae	Polyanthes tuberosa	Tube rose	13
2	,,	Amaryllis sp.	Easter-lily	8
3	.,	Hymenocallis sp.	Spider-lily	13
4	Anacardiaceae	Mangifera indica	Mango	13
5	Anonaceae	Anona squamosa	Custard apple	10
6	Araceae	Syngonium sp.		10
7	Begoniaceae	Begonia sp.	_	17
8	Bromeliaceae	Ananas comosus	Pineapple	13
9	Cannaceae	Canna indica	_	20
10	Caricaceae	Carica papaya	Pappaya	17
11	Chenopodiaceae	Beta vulgaris	Beet root	17
12	Commelinaceae	Tradescantia fluminensis	pp-many	10
13	,,	Zebrina pendula		17
14	Compositae	Helianthus annuus	Sunflower	10
15	Cruciferae	Brassica juncea	Mustard	9
16	,,,	B. oleracea	Cabbage	10
17	Cucurbitaceae	Cucurbita maxima	Pumpkim	8
18	,,	Cucumis sativus	Cucumber	12
19	,,	Citrullus vulgaris	Water melon	7
20	Euphorbiaceae	Ricinus communis	Castor	12
21	**	Codiaeum variegatum	Croton	10
22	,,	Manihot utilissima	Tapioca	17
23	Graminaceae	Oryza sativa	Rice	18
24	**	Eleusine coracana	Ragi	8
25	.,	Triticum vulgare	Wheat	9
26	**	Sorghum vulgare	Jowar	7
27	.,	Bambusa tulda	Bambo	7
28	,,	Zea mays	Maize	8
29	**	Saccharum officinarum	Sugarcane	24
30	Hydrocharitaceae	Vallisneria sp.		16
31		Hydrilla sp.		14
32	Labiatae	Mentha arvensis	Mint	6

(Contd.....)

\$. No. Family		Species	Common name	Max. No of days survived	
. 33	Leguminosae	Dolichos lah lah	Lab lab	8	
34	,,	Arachis hypogea	Groundnut	8	
35	.,	Pisum sativum	Pea	8	
36		Vigna sinensis	Cowpea	8	
37		Albizzia lebbek	_	8	
38	Liliaceae	Allium cepa	Onion	9	
39	Malvaceae	Abelmoschus esculentus	Bhendi	10	
40	,,	Gossypium arboreum	Cotton	21	
41	Moraceae	Artocarpus heterophyllus	Jack fruit	8	
42		Ficus carica	Fig	12	
43	Myrtaceae	Psidium guajaya	Guava	8	
44	Oleraceae	Jasminum nudiflorum	Jasmine	10	
45	Orchidaceae	Vanilla fragrans	Vanilla orchid	30	
46	Palmaceae	Cocos nucifera	Coconut	8	
47		Araca catechu	Beetlenut	8	
48	Parkeriaceae	Azolla pinnata	_	30	
49	Piperaceae	Peperomia sp.	_	8	
50	Punicaceae	Punica granatum	Pomegranate	6	
51	Rosaceae	Rosa alba	Rose	19	
<b>5</b> 2	Rubiaceae	Coffea robusta	Coffee	10	
53	Rutaceae	Citrus media	Lime	10	
54		Murraya exotica	Curry leaf	12	
5.5	Sapotaceae	Achras zapota	Sapota	9	
56	Scitamineae	Musa paradisiaca	Banana	8	
57	Solanaceae	Solanum tuberosum	Potato	9	
58	,,	S. melongena	Brinjal	8	
<b>5</b> 9	,,	Capsicum annum	Chilli	10	
60	**	Lycopersicon esculentum	Tomato	9	
61	Theaceae	Thea sinensis	Tea	7	
62	Umbelliferae	Coriandrum sativum	Coriander	10	
63	,,	Daues carota	Carrot	7	
64	Vitaceae	Vitis vinifera	Grape	10	
65	Zingiberaceae	Zingiber officinale	Ginger	7	
66		Curcuma longa	Turmeric	11	
67		Elettaria cardamomum	Cardamom	18	

S. No.	Family	Species	Common name	Max. No. of days survived	No. of feed- ing spots observed
1.	Araceae	Amorphophallus sp.	Yam	7	1
2.	**	Colocasia esculenta	Arvi	8	3
3.	,,	Pistia stratiotes	-	17	18
4.	Convolvulaceae	Ipomoea batatas	Sweet potato	28	5
5.	Compositae	Lactuca sativa	Lettuce	10	2
6.	Cruciferae	Raphanus sativus	Radish	25	5
7.	Nymphaceae	Nymphaea spp.	Water lily	9	3
8.	Onagraceae	Trapa bispinosa	Water chestnu	t 18	10

TABLE 2. Test plants on which nibbling by C. salviniae was observed.

complete development in any plant other than Salvinia. Subsequent tests carried out in Australia (FORNO et al., 1983) also confirmed this. It was therefore concluded that C. salviniae is incapable of reproducing on any of the test plants, other than S. molesta.

Permission of the plant protection Adviser to the Government of India was obtained in September 1983 for field trials and 785 adults were released in a Salvinia infested lily pond at the Lalbagh Botanical Gardens in Bangalore. Establishment of the insect under field conditions was observed by January 1984, as evidenced by the presence of larvae, pupae and freshly emerged adults. A nucleus culture of C. salviniae was also supplied to Dr. P. J. Joy of the Kerala Agricultural University at Trichur for breeding and subsequent field trials in Kerala. personal communication to the second author of this paper Dr. Joy (1985) has mentioned that C. salviniae is proving to be a great success and is now spreading to the vast stretches of Kuttanad in the Alleppey district of Kerala. In a subsequent study Room et al., (1984) found that establishment of C. salviniae could be achieved with the release of as few as 200 adults and that the shortest time for damage to be caused was 4 months. They also found that establishment occurred in areas where nitrogen concentration was 1.18-1.82% of dry weight of the weed and where temperatures ranged from 0-45°C. Establishment occurred in 7 of the 8 sites where releases were made.

The biological control efforts against Salvinia are a clear indication of the potential that this method of control holds for weed control in India. Moreover, it should help to allay fears of introducing insect natural enemies of weeds into the country, particularly when the natural enemy in question has been throroughly studied and proven to be safe to cultivated crops. Although it is too early to say that Salvinia control in India will be as spectacular as it was in Australia, there is reason to believe that the project is certainly going in that direction.

Acknowledgements. The authors are grateful to Dr. Harley of Long Pocket Laboratories, Australia, and to the Commonwealth Institue of Biological Control for their help in procuring stocks of C. salviniae and also to the latter

for the use of their quarantine facilities at Bangalore. They also acknowledge with gratitude the encouragement provided by the Indian Council of Agricultural Research and the Director, Indian Institute of Horticultural Research, Bangalore for facilities. Technical assitance given by Mr. S. K. Jalali is also acknowledged.

#### **RRFERENCES**

- Bennett, F. D. (1966) Investigations on the insects attacking the aquatic fern Salvinia spp. in Trinidad and northern and northern South America. Proc. S. weed Conf., 19, 497-504.
- CALDER, A. A. & D. P. A SANDS (1985) A new Brazilian Cyrtobagous Hustache (Coleoptera: Curculionidae) introduced into Australia to control salvinia. J. Aust. ent. Soc., 24, 57—64.
- Forno, I. W. (1981) Progress in the exploration for biological control agents for Salvinia. Proc. V Int. Symp. Biol. Contr. Weeds, Brisbane, Australia, 1980, 167-173.
- Forno, I. W. & K. L. S. HARLEY (1979) The occurrence of Salvinia molesta in Brazil. Aquatic Botany, 6, 185—187.
- Forno, I. W., D. P. A. Sands, & W. Sexton (1983) Distribution, biology and host-specificity of *Cyrtobagous singularis* Hustache (Coleoptera: Curculionidae) for the biolo-

- gical control of Salvinia molesta. Bull. ent. Res., 73, 85-95.
- Joy, P. J. (1978) Ecology and control of Salvinia (African Payal) the molesting weed of Kerala. Tech. Bull. No. 2, Kerala Agric. Univ., 40 pp.
- Joy, P. J., K. C. VERGHESE & C. C. ABRAHAM (1981) Studies on biology and host range of *Paulinia acuminata* De Geer (Orthoptera: Acrididae) and its efficacy for the control of *Salvinia molesta* Mitchell an aquatic floating weed in Kerala. *Proc. 8th Asian Pacific weed Sci. Soc. Conf.*, 201—206.
- Room, P. M., K. L. S. Harley, I. W. Forno & D. P. A. Sands (1981) Successful biological control of the floating weed *Salvinia*. *Nature*, 294, 78—80.
- Room, P. M., I. W. Forno & M. F. J. Taylor (1984) Establishment in Australia of two insects for biological control of the floating weed Salvinia molesta. Bull. Ent. Res, 74, 505—516.
- Sands, D. P. A. (1983) Identity of *Cyrtobagous* sp. (Coleoptera: Curculionidae) introduced into Australia for biological control of *Salvinia. J. Aust. Ent. Soc.*, 22, 200.
- Sankaran, T. & G. Ramaseshaiah (1973) Investigations on three species of insects recommended for biological control of the aquatic weed Salvinia auriculata. Unpubl. Rept., Commonw. Inst. Biological Contr., Indian Station, Bangalore, 6 pp.

# PROTEIN AND GLYCOGEN CONTENTS OF THE ACCESSORY REPRODUCTIVE GLANDS OF THE MALE AND FEMALE SILK MOTHS BOMBIX MORI BEFORE AND AFTER MATING

S. H. BHOSALE, V. L. KALLAPUR & K. VENKATESH\*

Department of Zoology, Karnatak University, Dharwad, India 580 003
\* Department of Entomology, North Carolina University, North Carolina, U S A

(Received 20 November 1985)

The accessory reproductive glands (ARGs) of the mated male silk moth show highest concentration of total protein when compared to the testis, vas deferens, seminal vesicle and ejaculatory duct. The protein level of the ARGs show significant depletion after mating. The gravid female silk moth shows significant increase of the protein content of the accessory reproductive structures after mating. It is believed from these observations that the male proteins are transferred to the female during copulation. Using <sup>14</sup> C-leucine, it is possible to demonstrate that the male transferred proteins are stored in the accessory reproductive structures of the mated female. Glycogen appears to be the chief source of energy during mating in the male, whereas it serves as source of energy during ovulation process in the female moth.

(Key words: Protein, glycogen, accessory male and female reproductive glands, silk moths, Bombyx mori)

#### **INTRODUCTION**

Several studies have shown that the proteins from the male accessory reproductive glands (ARGs) are transported during copulation (LEOPOLD et al., 1971; TERRANOVA et al., 1972; KAULENAS, 1976; CHEN, 1984). It is known that the male ARGs produce the proteins which are used in the assembly of spermatophore, a structure which serves as the vehicle for the transfer of sperm from male to female (Wigglesworth, 1936). from other insects have indicated that material transferred by the male to the female during copulation either in the form of spermatophore or as seminal secretions may be used as source of nutrient by the female (LEOPOLD, 1976). This is of particular importance to the

lepidopteran species which do not feed during adult life. The accessory secretion was found to stimulate oviposition in the female (CHEN, 1984) and was involved in the cytolysis of the female vaginal pouches which occur during copulation (LEOPOLD et al., 1971; TERRANOVA et al., 1972). Thus, the male accessory secretion transferred during copulation seems to have an important role in the regulation of the physiology of the female and may contain several stimulatory substances. The present study examines the protein content of the male and female ARGs of the silk moths B. mori before and after mating. The present opportunity was also availed of to study the glycogen level of the ARGs and its role as a source of energy during copulation.

#### MATERIALS AND METHODS

The adults of the silk moths B. mori (NB<sub>4</sub>D<sub>2</sub>) were obtained from laboratory culture. Male moths were placed with virgin females and as mating began, each pair was placed in a separate container. After 3 h copulation, some pairs were forcibly separated and the ARGs were carefully removed from the male and female moths for the determination of total proteins and glycogen contents. The remaining mated females were allowed to lay eggs. The ARGs of the female moth were removed after the completion of the egg laying. Likewise the ARGs of the unmated male and female moths were used for the estimation of proteins and glycogen.

The protein content of the ARGs was precipitated by the addition of 30% trichloro acetic acid (TCA) solution, followed by centrifugation at 3000g for 10 minutes at 20°C. The precipitate was washed twice with the same volume of TCA solution and dissoloved in 1ml of 0.1N NaOH. Known aliquots of this solution was used for the protein determination according to the method of Lowry et al. (1951) using bovine serum albumin (fraction IV, Sigma Chemical Company, USA) as reference standard. Incorporation of (U-14C) leucine into the ARGs was essentially similar to that described by FRIEDEL & GILLOT (1977). Five microliter of laballed leucine was injected into

each insect (specific activity, 350.0 mCi/m mole). After one hour of incubation time the radioactive males were allowed to mate with females for three hours after which some pairs were forcibly separated. Males and females were dissected under cold Bombyx saline (YAMAOKA, et al., 1971). The various reproductive structures were removed and processed for the determination of radioactivity in the protein according to the method described by MANS & NOVELLI (1961). The radioactivity was measured on Beckman Liquid Scintillation Counter. The efficiency of the counter for 14c was more than 90 per cent. The glycogen content of the ARGs was estimated following the precipitation of glycogen from a saturated solution of sodium sulfate with absoulte ethanol. Glucose was used as reference standard. The procedure followed was essentially similar to that described by SEIFTER et al. (1950).

#### RESULTS AND DISCUSSION

The results are presented in Tables 1—4. It may be seen from the results outlined in Table 1 that in the unmated male, the ARGs show the highest concentration of proteins (668.14  $\mu$ g) when compared to those of testis (390.17  $\mu$ g), vas deferens (215.23  $\mu$ g), seminal vesicle (163.41  $\mu$ g) and ejaculatory duct (219.04  $\mu$ g). It is also evident that the protein

TABLE	1.	Protein	content	of	the	various	reproductive	structures	of	the male
			silk	mo	th b	efore and	l after mating	ζ.		

Reproductive structure		$\mu_{g}$ / :	%	P		
		Unmated male	Mated male	reduction	value	
Accessory reproductiv	e					
gland	(7)	668.14 ± 49.4°	$444.92 \pm 24.23$	33.4	< 0.005	
Testis	(7)	$390.47 \pm 27.30$	$314.28 \pm 17.02$	19.51	< 0.05	
Vas deferens	(7)	$215.23 \pm 19.80$	$158.09 \pm 14.07$	26.54	< 0.05	
Seminal vesicle	(7)	$163.41 \pm 10.21$	$121.86 \pm 12.07$	25.42	< 0.05	
Ejaculatory duct	(7)	$219.04 \pm 15.83$	$170.14 \pm 19.21$	22.31	< 0.1	

<sup>\*</sup> The results indicate mean ± SE of seven experiments; number of insects used is indicated in parenthesis.

level of these reproductive structures is depleted after copulation (Tables 1 & 2). The ARGs show maximum depletion (33.4%) than those of testis (19.51%), vas deferens (25.54%), seminal vesicle (25.42%) and ejaculatory duct (22.31%). It is of considerable interest to note that the gravid female shows significant increase in the protein content of the accessory reproductive structures

(Table 2). Bursa copulatrix of the mated female shows as much as 475% increase while spermatheca and accessory gland exhibited respectively 177.5% and 30.51% increase in the protein level. It is possible that the increased level of proteins in the female may be due to the stimulation of protein synthesis by the male factor (s) transferred during copulation or the proteins of the male accessory

TABLE 2. Protein content of the accessory reproductive structures of the female silk moth during reproductive cycle.

			$\mu$ g / glan	d		
Accessory repro- ductive structure		Virigin female	Mated female before egg laying	increase	Mated female after egg laying	decrease
Accessory gland	(7)	892.53 ± 36.93	1164.91 ± 76.09 ( P<0.01 )	30.51	$394.77 \pm 49.66$ ( P<0.001 )	66.11
Bursa copulatrix	(7)	77.77 ± 15.97	$447.61 \pm 28.03$ ( P<0.001 )	475.6	$313.21 \pm 24.46$ ( P<0.001 )	30.02
Spermatheca	(7)	63.48 ± 5.31	$176.18 \pm 6.63$ ( P<0.001 )	177.5	$81.93 \pm 5.81$ ( P<0.05 )	53.49

The results indicate mean ± SE of seven experiments.

Table 3. Recovery of U 14c-leucine from the accessory reproductive structures of the unmated male, mated male and mated female of the silk moth B. mori.

Accessory reproductive	Radioactivity (		
structures	unmated male	mated male	Mated female
Testis	10,020*	6230	_
Vas deferens	7,710	5926	-
Male ARG	10,748	6634	-
Seminal vesicle	7,165	5400	_
Ejaculatory duct	7,768	4473	-
Female ARG	_	_	2,931
Bursa copulatrix	_	_	3,219
Spermatheca	_	_	2,476
Ovary	-	_	_

<sup>\*</sup> The resutls are mean of three experiments.

TABLE 4. Glycogen content of the accessory reproductive glands of male and female silk moth B. mori.

		μg glycogen / gland		
Experimental insect			P value	
Non-mated male	(7)	67.50 ± 4.98		
Mated male	(7)	$43.79 \pm 2.80$	<0.001	
Virigin female	(7)	$65.75 \pm 6.93$		
Mated female before egg				
laying	(7)	$61.25 \pm 7.10$	< 0.001	
Mated female after				
egg laying	<b>(</b> 7)	$38.31 \pm 0.59$		

The result indicates mean  $\pm$  SE of seven experiments; number of insects used in indicated in parenthesis.

reproductive structures that are transferred may be the source of additional protein that occurred in the female accessory It may be of reproductive structures. particular interest to mention in this context that the male Cecropia moth transfers juvenile hormones (JH 1 & JH 2) to the female during copulation which are stored in the female ARGs (SHIRK et al., 1980). The fate of JH in the female is not known since it has been shown that vitellogenin synthesis in this moth is not influenced by JH. The results presented in Table 3, however, indicate that the male silk moth injected with 14C leucine when allowed to mate with the female, the accessory reproductive structures of the female showed accumulation of labelled protein. It is suggested that the proteins from the male are transferred during copulation in the silk moth B. mori. The role of these proteins, however, is unclear.

It is evident from the results presented in Table 2 that the protein content of the accessory reproductive structures of the female show substantial depletion of proteins after egg laying. Maximum depletion occurs in the ARG (66.11%) than bursa copulatrix (30.02%) and spermatheca (53.49%). In the silk moth *B. mori* the bursa copulatrix receives sperms during copulation and they are temporarily stored in the spermatheca. It appears that the increase of proteins in the gravid female and their depletion after egg laying may be due to the accumulation and utilization of transferred sperms for fertilization. Similarly, the decrease in the protein content of the female ARG may be due to their utilization for the egg coating (TAZIMA, 1978).

It is accepted that the glycogen serves as source of energy for various activities of insect (STEELE, 1982). The present study has revealed that the glycogen content of the male ARGs depleted significantly after mating. In the female, on the other hand the glycogen level is reduced substantially only after the egg laying. It seems that the glycogen is utilized as a sourcee of energy during mating in the case of male, whereas in the female it serves as source of energy during oviposition process.

#### REFERENCES

- CHEN, P. S. (1984) The functional morphology and biochemistry of insect male accessory glands and their secretions. A. Rev. Ent., 29, 233—255.
- FRIEDEL, T. & C. GILLOTT (1977) Contribution of male produced proteins to vitellogenesis in *Melanoplus sanguinipes*. J. Insect Physiol., 23, 145—151.
- KAULENAS, M. S. (1976) Regional specialization for export protein synthesis in the male cricket accessory gland. J exp. Zool., 195, 81-96.
- LEOPOLD, R. A., A. C. TERRANOVA, B. J. THORSON & M. E. DEGRUGILLIER (1971)

  The biosynthesis of the male housefly accessory secretion and its fate in mated female. J. Insect Physiol., 17, 987—1003.
- LEOPOLD, R. A. (1976) The role of male accessory glands in insect reproduction.

  A. Rev. Ent., 21, 199-221.
- LOWRY, L. H, N. J. ROSEBROUGH, A. L. FARR & R. J. RANDALL (1951) Protein measurement with the folin phenol reagent. J. biol. Chem., 2193, 65-275.
- Mans, R. J. & G. D. Novelli (1961) Measurements of the incorporation of radioactive amino acids into protein by a filter-paper disk method. Arch. Biochem. Biophys., 94, 48-53.

- SEIFTER, S., S. DAYTON, B. NOVIC & E. MUNTWLYER (1950) The estimation of glycogen with the anthrone reagent. Archf Biochem., 25, 191-200.
- STEELE, J. E. (1982) The role of carbohydrate metabolism in physiological function, 1—58, in: Energy Metabolism in Insects (Ed. R. G. H. DOWNER,) Plenum Publishing Corporation.
- SHIRK, P. D., G. BHASKARAN & H. ROLLER (1980) The transfer of juvenile hormone from male to female during mating in the *Cecropia* silk moth. *Experientia*, 36, 682-683.
- TAZIMA, Y. (1978) The silkworm an Important. Tool 30, Kodansha Tokyo, Japan.
- TERRANOVA, A. C., R. A. LEOPOLD, M. E. DEGRUGILLIER & J. R. JOHNSON (1972) Electrophoresis of the male accessory secretion and its fate in the matcd female. J. Insect Physiol., 18, 1573—1591.
- YAMAOKA, K., M. HOSHINO & T. HIRAO (1971) Role of sensory hairs on the anal papillae in oviposition behaviour of *Bombyx mori*. J. Insect Physiol., 17, 897—911.
- WIGGLESWORTH, V. B. (1936) The function of the corpus allatum in the growth and reproduction of *Rhodnius prolixus*, *J. exp. Zool.*, 79, 91-122.



# EFFECT OF CATHARANTHUS ALKALOIDS ON THE REPRODUCTIVE PERFORMANCE OF THE HOUSE FLY, MUSCA DOMESTICA L.

#### KUMUDA SUKUMAR

Pesticide Division, Regional Research Laboratory, Hyderabad, India

(Received 21 February 1986)

The total leaf alkaloids and root alkaloids of *Catharanthus roseus* induced significant sterility in males and females of the house fly, *Musca domestica*. The leaf alkaloid showed superior action when compared to root alkaloids. The root alkaloid, though inferior in its efficiency in inducing a higher degree of sterility, presented sex specificity by being more effective in females than males. When both sexes were treated, the alkaloids gave a cumulative effect. The plant deserves further attention since it shows commercial promise.

(Key words: Catharanthus alkaloids, Musca domestica, housefly, sterility)

#### INTRODUCTION

Natural products have always fascinated biologists because of their propensity to be used either as drugs or as insect control agents, and hence plant kingdom has been a fruitful hunting ground for bioactive compounds and many plants have emerged as important agents with significant bioactivity. Catharanthus roseus (Apocyanaceae) gained considerable reputation in recent years, since the discovery of anti-cancer properties of its alkaloids (NOBLE et al., 1958). Research on anticancer agents has provided many leads to the discovery of insect reproductive inhibitors (Borkovec, 1962). The majority of known reproductive inhibitors, reduce or eliminate the reproductive capacity of the insects and function mostly by attacking the rapidly proliferating cellular systems of the reproductive organs. Although a direct relationship has not been established between the two biological activities (CHANG et al., 1974) many anti-cancer agents have led to the development of an insect sterilant. The rapidly dividing cells of the insect reproductive system bear in some respect a close resemblance to those in a growing tumour and it is conceivable that a compound effective in one system could also affect the other. Sukumar & Osmani (1981) reported that the leaf and root alkaloids of Catharanthus held promise as good sterilants against Dysdercus cingulatus. The present work represents an extention of the study to Musca domestica to understand the generality of action of the alkaloids.

#### MATERIALS AND METHODS

Musca domestica was reared in the laboratory on a semisynthetic diet at  $27 \pm 3^{\circ}$ C. The pupae were removed from the culture medium and kept for emergence. Flies less than 24 h old, were sexed and used for treatment.

The total leaf alkaloids were extracted from air-dried leaves with methanol in a soxhlet apparatus, methanol removed and the residue treated with chloroform. The chloroform soluble portion was treated with 1% HCl

and to the acid soluble portion 20% ammonia added (pH 9.0) (JOSHI & AMBAYE, 1968). The total alkaloid was obtained as a gift from the Central Institute of Medicinal and Aromatic Plants, Bangalore, India.

The alkaloids were dissolved in chloroform and graded concentrations made. The desired concentrations of alkaloid solutions were applied topically on the dorsal side of the flies at the rate of 1  $\mu$ 1/insect. The treated insects were caged with the untreated, unmated, freshly emerged flies of the opposite sex and the ratio of the male to the female was maintained at 20:15. In all the experiments, the insects were allowed to mate and after oviposition, the eggs were collected at random. Three hundred eggs in three batchs of hundred each were kept in culture medium separately, for viability studies. The maggots which hatched were counted and the sterility recorded. All the experiments were conducted in triplicate and continued for 20 days. Check insects received only solvent applications. The mean value of the triplicate testing was taken as the percentage observable sterility. This was corrected with natural sterility using Abbott's formula (ABBOTT, 1925).

#### RESULTS AND DISCUSSION

The results show that Catharanthus alkaloids can successfully sterilize male and female of house flies. Table 1 shows the sterility data when the 1st batch of eggs in all the triplicate testing is taken into account. As in the case of Dysdercus the leaf alkaloid gave better sterilant

action than the root alkaloid. Svoboda (1963) and Svoboda et al. (1975) reported that the activity of the plant was found entirely in its alkaloidal constituents and the leaf alkaloids were far more active than those of stems or roots.

The root alkaloid, though generally inferior to the leaf alkaloid in its efficiency, presented a sex specificity by inducing a higher degree of sterility in females than in males. When both sexes were treated the alkaloids gave a cumulative effect. But it was also observed that treatment of both sexes with lower doses resulted in a progressive recovery from the sterilant action since the subsequent batches presented lesser sterilant action and the viability of eggs increased with passage of time (Table 2). Although the recovery was quite remarkable at lower doses, at higher doses like 2% the recovery was negligible. Most of the doses like 3% to 4% alkaloids permanently sterilized the insect giving little scope for recovery. This indicates the possibility of sterilizing the insects permanently with higher doses and preventing them from recovering from the sterilized effect. Mating was not inhibited at any dose in The plant deserves deeper either sex. investigation as it shows potential promise for commercial exploitation.

TABLE 1. Sterility data in Musca domestica following topical treatment with Catharanthus alkaloids.

Total leaf alkaloid				Total root alkaloid		
-0/ Dana	Percentage sterility		0/ D	Percentage sterility		
% Dose _	male	female	% Dose	male	female	
0.5	85.0	80	0.5	23	34	
1.0	87.5	85	1.0	32	40	
2.0	95.0	92	2.0	69	74	
			3. <b>0</b>	76	88	

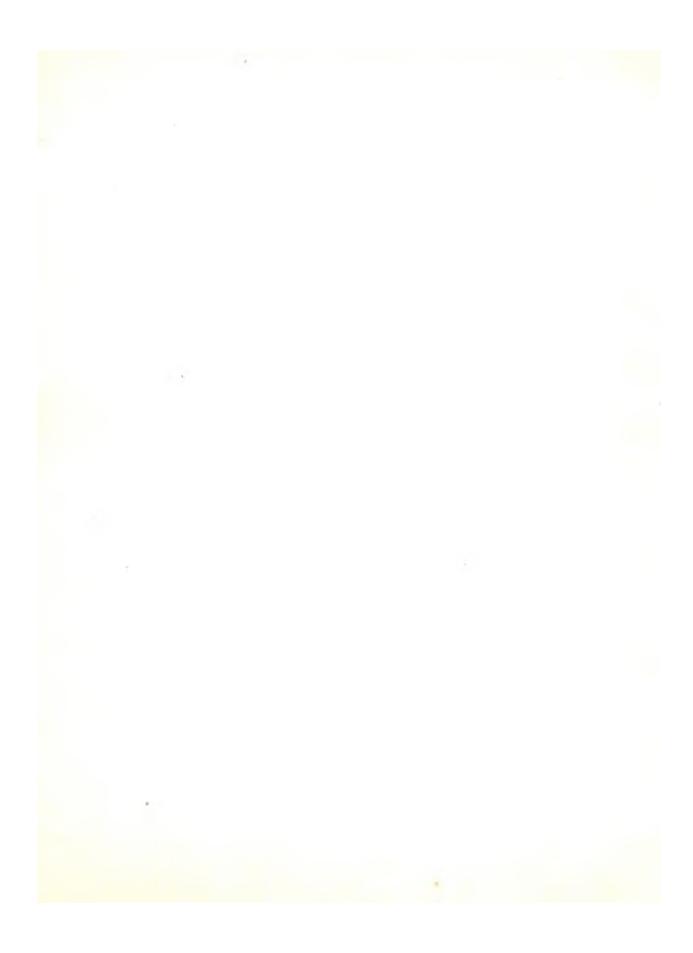
TABLE	2.	Recovery				following sca domesti	•	treatment	of	Catharanthu	5
do se	9/	sterility in	n 3 bai	ches of eg	g <b>g</b> s	% dose	% S1	terility in 3	ba	tches of egg	s

% do se leaf al-	% sterility in 3	batches of eggs	% dose	% sterility in	3 batches of eggs
kaloid	treated male	treated female	alkaloid	treated male	treated female
0.5	I Batch = 85	I Batch = 80	0.5	I Batch = 23	I Batch $= 34$
	II Batch = 58	II Batch $= 60$		II Batch = 17	II Batch = 21
	III Batch = 49	III Batch = 43		III Batch ≈ I2	III Batch = 7
1.0	I Batch = 87	I Batch $= 85$	1.0	I Batch = 32	I Batch $= 40$
	II Batch $= 65$	II Batch $= 63$		11 Batch = 26	II Batch = 28
	III Batch = 57	III Batch = 52		III Batch = 14	III Batch = 19
2.0	I Batch $= 95$	I Batch = 92	2.0	I Batch $= 69$	I Batch $= 74$
	II Batch $= 93$	IJ Batch = 89		II Batch = 66	II Batch = 70
	III Batch $= 90$	III Batch = 87		III Batch = 60	III Batch $= 65$

#### REFERENCES

- ABBOTT, W. S. (1925) A method of computing the effectiveness of an insecticide. *J. econ. Ent.*, 18, 265—267.
- BORKOVEC, A. B. (1962) Sexual sterilization of insects by chemicals. Science, 137, 1034—1037.
- CHANG, S. C., A. B. BORKOVEC & B. H. BRAUN (1974) Chemosterilant activity of antineoplastic agents. Trans. N. Y. Acad. Sci., 36, 101-107.
- NOBLE, R. L., C. T. BEER & J. H. CUTTS (1958) Role of chance observations in chemotherapy: Vinca rosea. Ann. N. Y. Acad. Sci., 76, 882-894.

- SUKUMAR, K. & Z. OSMANI (1981) Insect sterilants from Catharanthus roseus. Curr. Sci., 552—553.
- SVOBODA, G. H. (1963) Alkaloids of Vinca rosea Linn. (Catharanthus roseus G. Don) root alkaloid. J. Pharm. Soc., 18(52), 407—408.
- SVOBODA, G. H. & D. A. BLAKES (1975) The phytochemistry and pharmacology of Catharanthus roseus (L), 45-84, in: The Catharanthus alkaloids: (Ed. W. TAYLOR & N. FARNSWORTH) New York: Marcel Dekker Inc.
- Joshi, M. S. & R. Y. Ambaye (1968) Effect of alkaloids from *Vinca rosea* L on spermatogenesis in male rats. *Ind. J. exp. Biol.*, 6, 256—257.



# EFFECTIVENESS OF SPRAY FORMULATIONS AGAINST RICE LEAF FOLDER CNAPHALOCROCIS MEDINALIS GUENEE (LEPIDOPTERA: PYRALIDAE)\*

MANGAL SAIN, N. V. KRISHNAIAH & M. B. KALODE

Directorate of Rice Research, Rajendranagar, Hyderabad, India 500 030

(Received 24 October 1985)

Fifteen insecticides available in the market were tested as high volume sprays while eight selected insecticides were evaluated as low volume sprays against leaf folder. Results revealed monocrotophos, chlorpyriphos, quinalphos and methyl parathion as effective insecticides.

(Key words: insecticides, spray formulations, leaf folder, rice, control with insecticide sprays)

#### INTRODUCTION

Earlier studies on chemical control of rice leaf folder, Cnaphalocrocis medinalis Guenee identified parathion or a mixture of toxaphene and DDT as foliar sprays (BALASUBRAMANIAN et al., 1973); methyl parathion, phenthoate and endosulfan spray (Das & Nair, 1975); chlordimeform granules and spray (VELUSAMY et al., 1978); carbaryl and methyl parathion (RAI & VIDYACHANDRA, 1979) as effective insecticides. Recently chlorpyriphos methyl and cartap (ENDO & MASUDA 1981) as well as thiocyclam hydrogen oxalate, isofenphos and carbosulfan (RAZVI et al., 1983) were found to be better insecticides against this pest. Among the synthetic pyrethroids permethrin was particularly efficient in reducing leaf damage (SAROJA & RAJU, 1982b). Present studies were carried out to generate comprehensive information on the effectiveness of marketed spray formulations against leaf folder under field conditions.

#### \* DRR Publication No. 279

#### MATERIALS AND METHODS

Fifteen insecticide formulations available in the market were included in evaluation as high volume sprays (Table 1) while eight selected insecticides were evaluated as low volume sprays (Table 2). The insecticides effective against other rice insect pests were given preference for inclusion as low volume sprays while the number depended on the availability of experimental area.

In experiment with high volume sprays the plot size was 12 sq m with three replication while in low volume sprays the plot size was 44 sq m with 4 replications. All the insecticides were tested @ 0.5 kg ai/ha. In case of high volume spray the quantity of water required was 1000 litres/ha while in case of low volume spray 150 litres/ha was sufficient. Two sprays at 11 days interval were given in both the experiments. Observations on the leaf folder damaged leaves were taken one day before first spray application and twenty one days later. Only green damaged leaves were considered for estimation of damage. The leaves with 15 per cent or more damaged area were considered as damaged leaves. Five days after first spray 100 folded leaves from each plot were collected and dissected for number of larvae alive or dead and percentage mortality was calculated.

TABLE 1. Effectiveness of spray formulation against leaf folder as high volume sprays.

Insectio	ide	Formu-	ADL	/Hill	% larval*
Common name	Trade name	lation	(65 DAT)	AT (87 DAT)	mortality (70 DAT)
Quinalphos	Ekalux	25 EC	17.5	4.5	71 (84)
Thiometon	Ekatin	25 EC	17.8	6.2	15 (7)
Chlorpyriphos	Dursban	20 EC	16.6	5.8	59 (73)
Phosalone	Zolone	35 EC	17.6	6.3	21 (19)
Monocrotophos	Nuvacron	40 EC	18,6	6.2	78 (89)
Phosphamidon	Dimecron	40 EC	16.0	5.3	25 (18)
Fenthion	Lebaycid	100 EC	14.2	5.9	47 (54)
Endosulfan	Hildan	35 EC	14.9	6.4	44 (48)
Fenitrothion	Sumithion	50 EC	14.2	6.0	49 (57)
Methyl parathion	Metacid	50 EC	15.1	9.2	55 (67)
Malathion	Malathion	50 EC	15.4	12.2	35 (34)
Dichlorvos	Nuvan	100 EC	12 1	8.3	19 (16)
Carbaryl	Sevin	50 WP	13.6	8.9	19 (15)
Carbaryl	Svimol	40 LVC	14.4	9.8	31 (27)
Dimethoate	Rogor	30 EC	14 4	21.2	14 ( 6)
Untreated control			15.1	13.6	20 (12)
C D (0.05)			NS	2.3	21
C V (%)			20.1	16.3	33.4

<sup>\*</sup> The values in parentheses are original values. NB: All the insecticides were applied @ 0.5 kg ai/ha. ADL = Average damaged leaves. BT = 1 day before first treatment; AT = 21 days after first treatment.

#### RESULTS AND DISCUSSION

Considering the data on both leaf damage and larval mortality from both the experiments (Tables 1 & 2), monocrotophos, chlorpyriphos, quinalphos and methyl parathion could be judged as better insecticides against rice leaf folder. Methyl parathion, monocrotophos, quinalphos (Das & Nair, 1975) and methyl parathion (Rai & Vidyachandra, 1979) were reported to be effective against leaf folder which corroborate with the present findings. On the other hand, carbaryl (Rai & Vidyachandra, 1979), fenitrothion,

phosphamidon and dimethoate (Das & Nair, 1975) though reported to be effective insecticides did not exercise satisfactory control in the present investigation. Among the new spray formulathiocyclam hydrogen isofenphos and fenvalerate registered good degree of effectiveness (RAZVI et al., 1983), while SAROJA & RAJU (1982a) reported bendiocarb and acephate as the most effective insecticides against this Further it is evident that low pest. volume (LV) spraying was equally effective to high volume spraying. Therefore, low volume spraying could be used

Insectio	ide	Formu-	AD:	L/ <b>H</b> ill	% larval*
Common name	Trade name	lation	BT (57 DAT)	AT (79 DAT)	mortality (62 DAT)
Quinalphos	Ekalux	25 EC	11.6	8.2	58 (66)
Thiomaton	Ekatin	25 EC	10.3	7.3	38 (39)
Chlorpyriphos	Dursban	20 EC	9.6	6.5	81 (92)
Phosalone	Zolone	35 EC	9.2	10.3	42 (40)
Monocrotophos	Nuvacron	40 EC	10.3	6.1	75 (87)
Endosulfan	Hildan	35 EC	10.6	10.5	42 (46)
Methyl Parathion	Metacid	50 BC	11.7	6.7	81 (92)
Carbaryl	Sevimol	40 LVC	11.8	<b>9</b> .9	25 (18)
Untreated control			12.3	14.1	18 (13)
C D (0.05)			NS	2.15	32
C V (%)			14.8	16.7	42.8

Table 2. Effectiveness of selected spray formulations against leaf folder as low volume sprays.

to control leaf folder because of its obvious advantages of covering more area per unit time and less traversing per unit coverage. It was estimated that one hectare area could be covered within 8.3 hours by low volume spray as compared to 31.3 hours required for high volume spray.

#### REFERENCES

BALASUBRAMANIAN, G., M. SARVANABHAVANANDAM & T. R. SUBRAMANIAM (1973) Control of rice leaf roller *Cnaphalocrocis medinalis* Guenee. *Madras agric. J.*, 60(7), 425-427.

Das, N. M. & M. R. G. K. Nair, (1975) Relative contact toxicity of insecticides to the caterpillars of *Cnaphalocrocis medinalis* Guenee. *Agric. Res. J.*, *Kerala*, 12(2), 209-212.

Endo, S. & T. Masuda (1981) Evaluation of insecticides applied to rice plant for control of rice leaf roller Cnaphalocrocis me-

dinalis Guenee. Proceedings of the Assoc. for Plant Protection of Kyushu, 27, 87-89.

RAI, P. S. & P. VIDYACHANDRA (1979) Efficacy of carbaryl and methyl parathion for the control of *Cnaphalocrocis medinalis* (Guenee) (Lepidoptera, Pyralidae), infesting rice. *Current Research*, 8(12), 213—214.

RAZVI, S. A., C. S. RAO & N. V. RAO (1983) Efficacies of new granular and spray insecticides on rice pests. *Int. Rice Res. Newsletter*, 8(1), 15-16.

SAROJA, R. & N. RASU (1982a) Effect of foliar insecticides on stem borer and leaf folder. *Int. Rice Res. Newsletter*, 7(3), 14.

SAROJA, R. & N. RAJU (1982b) Synthetic pyrethroids for controlling leaf folders on rice. Int. Rice Res. Newsletter, 7(2), 15.

Velusamy, R., I. P. Janaki, & S. Jayaraj (1978) Efficacy of certain insecticides as granular and foliar formulations in the control of stem borer, leaf roller and planthopper in rice. *Pesticides*, 12(2), 11—14.

<sup>\*</sup> The value in parentheses are original values. NB: All the insecticides were applied @ 0.5 kg ai/ha. ADL = average damaged leaves; BT = 1 day before first treatment; AT = 21 days after first treatment.



### DAY BITING MOSQUITOES (DIPTERA: CULICIDAE) OF MANIPUR

K. B. RAJPUT<sup>1</sup> & T. K. SINGH

Department of Life Science, Manipur University, Manipur, India 795 003

(Received 31 July 1985)

A total of twenty three spacies of mosquitoes with day biting habit belonging to six genera, viz Anopheles, Aedes, Armigeres, Heizmania, Mansonia and Tripteroides were recorded from the State. Monthly fluctuation in day biting density of day biters have been presented for Manipur Valley.

(Key words: day biting, mosquitoes, Manipur)

#### INTRODUCTION

The host-vector contact plays a pivotal role in the dynamics and probability of disease transmission by the vector species. The probability of infestation, and intensity increases with the increasing number of biting by the vector species. This probability of infestation definitely increases for the vector species which use to bite during day also. In diurnal species, the availability of host during day in the area also intensifies the probability of host-vector contact. In the forested or industrial dumping areas these day biters bacome very much of an annoyance to human beings.

In view of above, the present study was conducted during October, 1983 to September 1984 to know the day biting mosquitoes of the State. During the study regular fortnightly sampling from valley and erratic survey with emphasis on day biting species was done from some hilly regions of the State.

#### MATERIALS AND METHODS

Fortnightly human bait collections during day for one hour in each fortnight in the Oak (Qurcus serrata Thunb.) plantation at Regional Tasar Research Station, Imphal (Manipur) was maintained for one year. For the bait collection self bait with aspirator tube collection technique was applied (W HO, 1975). Some erratic samples were also taken from the hilly region of the State from varied topographic divisions. The mosquitoes collected were killed with ether and kept seperately with date and locality record for identification. The collected specimens were identified with the key of BARRAUD (1934), CHRISTOPHERS (1933), Catalog of the KNIGHT & STONE (1977), and RAO (1984). The specimens were preserved and includes in the collection of Life Science Department, manipur University Manipur (India).

#### **RESULTS**

A total of 365 mosquitoes belonging to 6 genera and 23 species, were collected during biting in day. Out of these 9 species, viz: Anopheles barbirostris, Aedes albopictus, Ae. caecus, Ae. vexens, Ae. andamanensis, Ae. lineatopennis, Armigeres subalbatus, Mansonia indiana and M. uniformis were collected from valley of the State and some 18 species (Table 1)

Present address: Regional Tasar Research Station, Mantripokhari, Imphal, Manipur, India 795 002.

#### K. B. RAJPUT AND T. K. SINGH

TABLE 1. Day biting mosquitoes from hilly region of Manipur.

S. No	Species	No. of females collected	Locality	*Tope- graphy
1	Anopheles barbirostris Vander-			
	wulp, 1884.	01	Geljang (850m)**1, 27.xii.84.	1
2	Aedes albopictus (Skuse), 1894	09	Moltam (850m) 1, 1.xi.84	1
			Moreh (150m) 2, 18.xii.84	2
			Nungba (759m) 8.x.84	3
3	Ae. mediopuctatus Theobald, 1905	07	Nungba, 6, 8.x.84	3
			Tamenglong (12 room) 1, 8.ix.'84	4
4	Ae. formosensis Yamada, 1921	02	Nungba, 2, 8.x.84	3
5	Ae. niveus group	04	Nungba, 4, 8.x.84	3
6	Ae. (Finlaya) sp.?	01	Moltam, 1, 1.xi.84	1
7	Armigeres subalbatus	21	Jiribam (850M) 15, 7.x.84	5
			Moreh 6, 18. viii. 84	2
8	Ar. theobaldi Barraud, 1933	05	Moreh, 5, 18.viii.84	2
9	Ar. dentatus Barraud, 1927	05	Nungba, 5, 8,x.84	3
10	Ar. digitatus Edwards, 1914	16	Nungba, 16, 8, x.84	3
<b>‡</b> 1	Ar. flavus (Leicester) 1908	01	Nungba, 1, 8.x.84	3
12	Ar. inchoatus Barraud, 1927	03	Nungba, 3, 8.x.84	3
13	Ar. longipalpis (Leicester) 1904	05	Nungba, 3, 8.x.84	3
			Tamenglong 2, 8.x.84	4
14	Ar. magnus (Theobald) 1908	02	Nungba, 2, 8.x.54	3
15	Ar. omissus Edwards, 1914	34	Nungba. 34, 8.x.84	3
16	Heizmania complex (Theobald) 1910	12	Moreh, 6, 18.viii.84	2
			Nungba, 6, 8.x.84	3
<b>1</b> 7	Mansonia uniformis (Theobald) 1901	01	Moltam, 1, 1,xi.84	1
18	Tripteroides powelli var indicus Barraud, 1929.	01	Nungba, f, 8.x.84	3

130

<sup>1.</sup> Hilly region, with prevalent oak forest. (Qurcus serrata and Q. dealbata).
2. Hilly region with dense tree forest. 3. Hilly region with predominent bamoo bushes.

<sup>4.</sup> Hilergil yon with mixed type of forest. 3. Plain region, with domestic habitations.

<sup>\*\*</sup> M. S. L. Altitude of the place.

were collected from hilly region. Ano-Aedes pheles barbirostris, albopictus. Armigeres subalbatus and Mansonia uniformis were common from both hilly and valley region. The most important day biters of the State may be pointed out as Armigeres subalbatus, Aedes albopictus, Armigeres omissus, Aedes vexens, Mansonia indiana, M. uniformis, Heizmania complex and Aedes mediopunctatus.

The annoyance of day biters in valley initiates wef June to November, with the peaks in July-August and November

(Fig. 1). No biting were recorded during the months of December and January probably due to being coldest months for the State.

Erratic surveys from different parts of the hilly region of the state reveals a very rich day biting fauna in the hilly region. During survey a number of day biters were collected especialy from predominent bamboo areas. It is noteworthy to mention that a single half hour self bait collection from Nungba forest yielded 13 species. In the forest, most of the day biters use to breed in bamboo cuts.

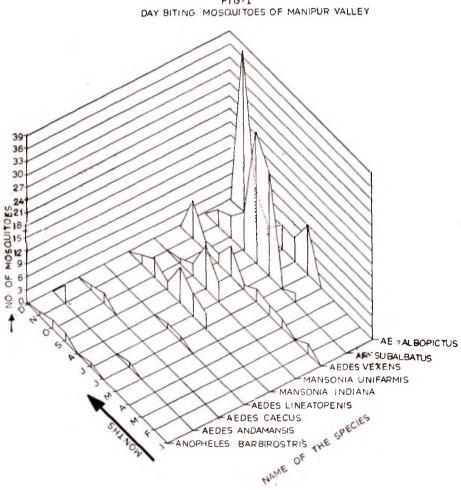


FIG-I

#### DISCUSSION

Recorded twenty three day biting mosquito species from the state belong to six genera viz., Anopheles, Aedes, Armigeres, Heizmania, Mansonia and Tripteroides. The maximum number of day biting species were recorded from genera Aedes and Armigeres (9 species each) followed by Mansonia, (2 species) Anopheles, Heizmania and Tripteroides (1 species each).

Among anophelines Anopheles barbirostris which is a reported secondary vector of human filariasis in India (RAGHAVAN, 1969), and found to harbour Japanese encephalitis virus from West Bengal (NIV. Document, 1979), was recorded to bite during day also in the jungles of the State. The similar day biting observations were also reported earlier by Christophers (1933), from Andaman Island and by Khin-Maung-Kyi (1977), from adjoining Burma.

Genus Aedes which contains 9 daybiting mosquito species represents Aedes albopictus. Ae. vexens (in valley) and Ae. mediopunctatus (in bamboo predominent areas) serves as severe day biters, while other species were not so annoying. Except the predominent species Aedes albopictus recorded from both hilly and valley regions, the two regions represent its seperate day biting aedine fauna. Aedes vexens, Ae. caecus, Ae. and amanensis and Ae. lineatopennis were recorded from valley while Ae. mediopunctatus, Ae. formosensis, Ae. niveus group and Ae. (Finlaya) sp. were recorded from hilly region of the State. Aedes albopictus a reported vector of Dengue fever in Singapore (CHAN et al., 1964), vector of Dinofilaria immitis in Nagasaki (Japan) and Thailand (HORSFALL, 1972), and capable of transmitting chickungunya virus in laboratory trials (RAO et al. 1964), maintains its second position in the State as day biter.

Genus Armigeres which includes the most vicious biter of the State, Armigeres subalbatus is prevalent in valley and hilly regions of the State. In valley Ar. subalbatus which is mainly domestic discarded container breeder, maintains its first position as day biter while in the hilly areas with predominent bamboo bushes Ar. omissus (bamboo breeder) ranks first. The other bamboo breading members of the group viz: Ar. dentatus, Ar. digitatus, Ar. flavus, Ar. inchoatus, Ar. longipalpis and Ar. magnus shares the day biting activity in the hilly region.

Genus *Heizmania* represented by *Heizmania complex* was collected during day biting from hilly region with predominent bamboo bushes. The day biting behaviour of the species was unknown from the work presented by BARRAUD (1934).

Mansonia uniformis and M. indina which are the important filaria vectors of India (RAGHAVAN, Loc. Cit.) were recorded biting during day also from the state.

Under genus Tripteroides, Tridteroides powelli var. indicus was collected during day biting from bamboo forest. Observation "The species is diurnal but not abundant or troublesome' (BARRAUD, 1934) was also found correct during the present study

Acknowledgements. The authors are thankful to Dr. B. A. HARRISON, Manager, Walter Reed Biosystematics unit, National Museum of Natural History, Washington D. C. for kind confirmation of some mosquitoes. Sincere thanks are also due to Mr. M. P. NIGAM, Joint Director and Dr. D. Subba Rao, Scnior Research Assistant, Regional Tasar Research Station, Imphal for their help in various ways during work.

#### REFERENCES

- BARRAUD, P. J. (1934) The Fauna of British India including Ceylon and Burma. Vol V. (Taylor and Francis, London) 463 pp.
- Basio, R. G. & L. Santos-Basio (1974) On Philippine mosquitoes. XIV. Biting Cycles of some species in their natural forest habitat with particular reference to Aedes albopictus. Kulikasan, 3, 155—165.
- CHAN, Y. C., B. C. Ho & K. L. CHAN (1971) Aedes aegypti (L) and Aedes albopictus (Skuse.) in Singapore city. 5 observations in relation to dengue haemorrhagic fever. WHO, 44, 651—658.
- CHRISTOPHERS, S. R. (1933) The Fauna of British India Including Ceylon and Burma, Diptera Vol 4 Family culicidae, Tribe Anopheline (Taylor and Francis, London), 271 pp.
- HADDOW, A. J. (1960) Studies on the biting habits and medical importance of East African mosquitoes in the genus Aedes. I. Subgenera Aedimorphus, Banksinella and Dunnius. Bull. ent. Res., 50, 759-779.
- Haddow, A. J., ((1961) Studies on the biting habits and medical importance for East African mosquitoes in the genus Aedes II. Subgenera Mucidus, Diceromyia, Finlaya and Stegomyia. Bull ent. Res., 52, 317—351.

- Horsfall, W. R. (1972) Mosquitoes 'Their bionomics and relation to disease' Ist reprint. Hafner Publishing Company, New York.
- KHIN-MAUNG-KYI (1971) The anopheline mosquitoes of Burma. Uni. Burma. Life. Sci., Part 1 to 4. 281—493.
- KNIGHT, K. L. & A. STONE (1977) A catalog of the Mosquitoes of the world (Diptera: Culicidae). (The Geo. W. King Company, Baltimore, Maryland 1202—1661.
- Anonymous (1979) India Japanese encephalitis information Document. National Institute of Virology, Pune, NIV Pune, Dabholkar Press, 47 pp.
- RAGHAVAN, N.G. S (1969) A review of epidemiology of filariasis in India. J. Com. Dis., 1, 153-194.
- RAO, T. R.; K. R. P. SINGH & K. M. PAVIR (1964) Laboratory transmission of an Indian strain of chickungunya virus. Curr. Sci.; 8, 235—236.
- RAO, T. RAMACHANDRA (1984) Anophelines of India. Malaria Research Centre (ICMR), Delhi.
- Anonymous (1975) Manual on practical entomology in malaria vector bionomics and organisation of anti-malaria activities Part I and II, WHO, Offset publication.



### GROWTH PATTERN OF CORCYRA CEPHALONICA PUPA AND EFFECT OF DIFLUBENZURON ON PUPAL WEIGHT<sup>1</sup>

M. Sriramulu<sup>2</sup> & K. N. Mehrotra

Division of Entomology, Indian Agricultural Research Institute, New Delhi 110 012

(Received 31 July 1985)

Growth pattern in normal and diflubenzuron treated pupa of *Corcyra cephalonica* was studied. The pupa continuously lost weight. Normal pupa lost weight by 0.59 mg/day. The loss in weight in diflubenzuron treated pupa was 0.75 mg/day/pupa. (Key words: growth, diflubenzuron, histolysis, xenobiotic, metabolism)

#### INTRODUCTION

The quantitative measurement of growth is based usually on weight. Although growth with respect to increase in weight in the larval stage has been reported (Hussain & Mathur, 1944; Davey, 1955; Church & Robertson, 1966; Mehrotra et al., 1972; Meisner et al., 1976; Sundaramurthy, 1977; Warthen & Uebel, 1980), there is a dearth of information on this aspect in pupal stage, as far as pupal growth is concerned.

AGRELL (1949) observed mutual change in weight of thorax and abdomen during pupal development of *Calliphora erythrocephala* mainly due to histolysis of larval fat body in the abdomen and histogenesis of imaginal organs in the insect. No change in weight during pupal development was reported in *Tribolium confusum* (CHAUDARY & LEMONDE, 1966) and *Drosophila melanogaster* (CHURCH & ROBERTSON 1966). In contrast to this, decrease in weight during pupal development has been reported in *Culex pipiens* (CHEN,

Therefore, the present study is concerned with the pupal growth pattern of *C. cephalonica* and the effect of diffubenzuron on it.

#### MATERIALS AND METHODS

The pupae of Corcyra cephalonica Stainton (Pyralidae: Lepidoptera) were obtained from a laboratory colony maintained on broken Sorghum bicolor (L.) grains. Technical grade diflubenzuron was a gift from Dr. A.B. BORKOVEC, Insect Chemosterilants Laboratory, United States

<sup>1958;</sup> ROZEBOOM & TWOHY, 1958), Aedes aegypti (LANG et al., 1965) and in diapausing pupae of Papilio zelicron (SIMS, 1983). Moulting is primarily a mechanism of growth. VAN DAALEN et al. (1972) reported that benzoylphenylureas affect the moulting in insects. The effect of diflubenzuron on the weight of larvae was observed in Musca domestica (ISHAAYA & CASIDA, 1974) and Spodoptera litura (SUNDARAMURTHY, 1977). These studies revealed that diffubenzuron decreased the rate of increase of weight in larval development. Though loss in weight during development in insecticide treated larvae was reported in literature, so far such data on pupal development is not available,

Part of the thesis submitted to P. G. School, I. A. R. I., New Delhi for award of Ph. D.

<sup>2</sup> Department of Entomology, College of Agril., R'nagar, Hyderabad

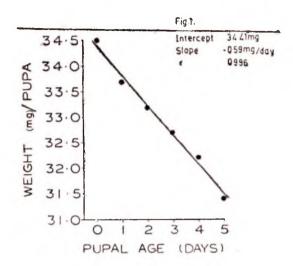
Department of Agriculture, Beltsville, Maryland, U. S. A.

To study the growth pattern, three replications of 20 pupae each were weighed at 24 h interval from the time of pupa formation till the emergence of adults. To study the effect of diflubenzuron treatment on pupal weight, 30 pupae were injected with 0.1 µg diflubenzuron per fresh pupa (0 day) and the pupae kept as control were injected with solvent only (dimethylformamide: methyl alcohol at 2:3 ratio) and were weighed at 24 h intervals throughout the pupal period. The pupae were injected with the help of an electrically operated micro-applicator (Model M 1025, Instrumentation Specialities Company, U.S.A.). The pupae of C, cephalonica were weighed with the help of a single pan balance type K-15, K-Roy, Varanasi, India.

#### **RESULTS**

Weight of pupa of C. cephalonica during development:

The data presented in Table I indicated that the insect continuously lost weight. The weight decreased from 34.5 mg to 31.4 mg during pupal development. When the data were subjected to linear regression analysis by the least squares



method, it gave a straight line (Fig. 1) with the regession equation Y = 34.41 - 0.59 X, where Y was the weight of pupa in mg and X the age of pupa. From the equation it would be seen that the pupa loses weight at the rate of 0.59 mg/day.

Effect of diffubenzuron on pupal weight during development:

From the data presented in the Table 2, it would be seen that in the solvent

Pupale age		mean weigh		
(days) —	R <sub>1</sub>	R <sub>2</sub>	R <sub>3</sub>	mg/pupa
0	693	696	679	34.5
1	681	676	667	33.7
2	672	662	660	33.2
3	662	647	654	32.7
4	650	634	649	32.2
5	637	618	629	31.4

TABLE 1. Weight of pupa of C. cephalonica during development.

R = Replication

For obtaining insects immediately on pupation the following method originally devised by de Wilde et al., (1968) was used. Larvae were released in petridishes containing teflon tubes. The larvae entered the tubes and pupated. These tubes containing light brown pupae were picked out and these were classified as 0-day pupae.

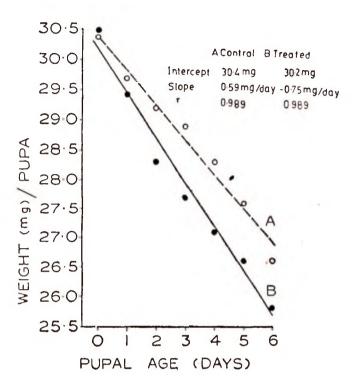


TABLE 2. Effect of diffubenzuron on pupal weight of C. cephalonica during development.

Pupal age	mean we	ight mg/pupa
(days)	control	treated
0	30.4	30.5
1	29.7	29.4
2	29.2	28.3
3	28.9	27.7
4	28.3	27.1
5	27.6	26.6
6	26.6	25.8

Each value is the mean 30 pupae.

Diffubenzuron (0.1  $\mu$ g/pupa) was injected into 0-day old pupa as a solution in 0.25  $\mu$ l of a mixture of dimethyl formamide and methanol (2:3 v/v). The controls were injected with solvent only.

treated control, weight decreased from 30.4 mg to 26.6 mg during pupal development period while it decreased from 30.5 to 25.8 mg in diflubenzuron treated pupa. These data, when subjected to linear regression by least squares method, gave a straight line (Fig. 2) with the regression equation Y = 30.4 - 0.59 X for control and Y = 30.2 - 75 X for diffubenzuron treated From the regression equations it could be seen that diflubenzuron treated pupa lost weight at the rate of 0.75 mg/day when compared to 0.59 mg/day in control pupa. The slope (-0.59 mg/day) of the solvent injected control pupa is similar to the slope of the normal pupa of the earlier experiment.

#### DISCUSSION

Although the growth pattern of larval stages have been extensively studied (Hussain & Mathur, 1944; Davey, 1955; CHURCH & ROBERTSON, 1966; MEHROTRA et al., 1972; WARTHEN & UEBEL, 1980), it has been poorly investigated in pupal stage. In C. cephalonica the pupa lost weight by 0.59 mg/day. Loss in weight during pupal development has been observed in Culex pipiens (CHEN, 1958; ROZEBOOM & TWOHY, 1958), Aedes aegypti (LANG et al., 1965). Absence of such loss of weight has been reported in Calliphora erythrocephala (AGRELL, 1949), Tribolium confusum (CHAUDHARY & LEMONDE, 1966) and melanogaster (CHURCH Drosophila ROBERTSON, 1966), The major events involved in the reorganisation during metamorphosis in insects are the histolysis of the tarval tissues and building up of the imaginal structures (CHEN, 1971). The nutrient reserves are the major energy source during metamorphosis. STRAUSS (1911) reported that the worker bee utilised 95 per cent of glycogen and 75 per cent of fat during the pupal period, CHEN (1958) reported a gradual decrease in free amino acids like alanine, leucine and  $\beta$  – alanine during pupal development of Culex pipiens. LANG et al., (1965) reported loss of water during pupal development of Aedes aegypti. Loss of water and utilisation of energy reserve for various metabolic processess during pupal development might be responsible for gradual decreases in pupal weight in C. cephalonica.

Diflubenzuron treatment increased the rate of loss in weight throughout the pupal period in *C. cephalonica* pupa. The loss in weight in treated pupa was 0.75 mg/day/pupa compared to 0.59 mg/day/pupa in normal insects. Although

there are no reports showing the loss of weight as a result of diffubenzuron or any other phenyl urea derivative in pupal stage, reduction in rate of increase in weight during larval development has been observed in other insects (ISHAAYA & Casida, 1974; Sundaramurthy, 1977). This loss in weight was most probably due to a higher rate of metabolism especially respiratory, induced by the insecti-The injected diflubenzuron is a xenobiotic and is to be metabolized and conjugated (CHANG, 1978; IVIE & WRIGHT, 1978) before it can be eliminated from the system. As the pupal stage in the holometabolousinsects is a closed system, diflubenzuron must be detoxified and stored in such a way that it does not affect the normal development. This detoxification mechanism also may increase metabolic activity. Enhanced metabolic activity would lead to a greater depiction of energy reserve probably resulting in more loss in pupal weight.

#### REFERENCRS

AGRELL, I. P. S. (1949) Localization of some hydrozen-activating enzymes in insects during metamorphosis. *Nature*, **164**, 1039—1040.

CHANG, S. C. (1978) Conjugation: The major metabolic pathway of 14c diflubenzuron in the housefly. J. econ. Ent., 71, 31-39.

CHAUDHARY, K. D. & A. LEMONDE (1966) In vivo synthesis and breakdown of deoxyribonucleic acid in Tribolium confusum Duval. Can. J. Biochem., 44, 1571-1575.

CHEN, P. S. (5958) Studies on the protein metabolism of *Culex pipiens* L. 1. Metabolic changes of free amino acids during larval and pupal development. *J. Insect Physiol.*, 2, 38-51.

CHEN, P. S. (1971) Biochemical Ascepts of Insect Development. S. Karger.

CHURCH, R. B. & ROBERSTON, F. W. (1966) A biochemical study of the growth of *Drosophila melangaster*. J. exp. Zaol., 162, 337—352.

- DAVEY, P. W. (1955) Quantities of food eaten by the desert locust, Schistocerca gregaria (Forsk.) in relation to growth. Bull. ent. Res., 45, 539-551.
- DE WILDE, J., G. E. STALL, C. A. D. DE KORT, A. DE LOOF & G. BOARD (1968) Juvenile hormone titre in the haemolymph as a function of photoperiodic treatment in the adult Colorado beetle (Leptinotarsa decemlineata Say). Proc. Kon. Ned. Akad. Westensch., Ser. 7, 321—326.
- Hussain, M. A. & C. B. Mathur (1944) Studies on Schistocerca gregaria Forsk. XI. The influence of temprature on growth in weight and size of the hoppers. *Indian J. Ent.*, 5, 107—115.
- ISHAAYA, I. & J. E. CASIDA (1974) Dietary JH 6040 alters composition and enzyme activity of housefly larval cuticle. *Pestic. Biochem. Physiol.*, 4, 484—490.
- LANG, C. A., H. Y. LAU & D. J. JEEFERSON (1965) Protein and nucleic acid changes during growth and aging the mosquito. Biochem, J., 95, 372—377.
- IVIE, G. W. & J. W. WRIGHT (1978) Fate of diffusenzuron in the stable fly and housefly. J. Agric. Food Chem., 26, 90—94.
- MEHROTRA, K. N., P. J. RAO, & T. N. A. FAROOQUI (1972) The composition, and uitilisation of food by locusts. *Ent. exp.* appl., 15, 91—96.

- Meisner, J., M. Wysoki & K., R. S. Ascher (1976) The residual effect of some products from neem (Azadirachta indica A. Juss) seeds upon larvae of Boarmia (Ascotis) selenaria Schiff. in laboratory trials. Phytoparasitica, 4, 185—192.
- ROZEBOOM, L. E. & D. W. TWOHY (1958) Comparison of nutritive reserves in males of autogenous and anautogenous populations of *Culex pipiens*. J. Parasitol., 44, 422—424.
- SIMS, S. R. (1983) Prolonged diapause and pupal survival of *Papilio zelteron* Lucas (Lepidoptera: Papilionidae). *J. Lepid Soc.*, 37, 29-37.
- STATUSS, J. (1911) Chemical composition during growth and metamorphosis. Apis. Z. Biol., 56, 347—97.
- Sundaramurthy, V. T. (1977) Effect on inhibitor of chitin deposition on the growth and differentiation of tobacco caterpillar Spodoptera litura Fb. (Noctuidae: Lepidoptera): Z. Pflkrantiz. Pflaschuttz.; 84, 597—601.
- VAN DAALEN, J. J., J. MELTZER, R. MULDER & K. WELLINGA (1972) A selective insecticide with a novel mode of action. Naturwissenschaffen, 59, 312—313.
- WARTHEN, J. D. J. & E. C. UEBEL (1930) Effect of Azadirachtin on house crickets, Acheta domesticus, Proc. Ist. Int. Neem Conj. Rottach. Egern, pp. 137—148.



# THE METABOLISM AND EXCRETION OF 14C-LABELLED DIELDRIN IN RESISTANT AND SUSCEPTIBLE TRIBOLIUM CASTANEUM (HERBST)

A. S. UDEAAN & R. L. KALRA

Department of Entomology, Punjab Agricultural University, Ludhiana-4, India

(Received 12 July 1985)

The metabolism and excretion of <sup>14</sup>C-dieldrin in the resistant and susceptible strains of *Tribolium castaneum* was studied. Adults of resistant strain were found to metabolize dieldrin to about six metabolites, two of which were identified as trans-aldrindiol and 9-hydroxy dieldrin. The metabolism of dieldrin in the resistant strain was more pronounced than in the susceptible strain. The high level of trans aldrindiol found in the resistant strain suggests that its formation was a detoxication reaction. The results also indicate the high rate of excretion of this insecticide by the adults of the resistant strain.

(Key words: metabolism, excretion, dieldrin, Tribolium castaneum)

#### INTRODUCTION

Tribolium castaneum (Herbst) has been reported to be resistant to lindane in various countries of the world, including India (CHAMP & DYTE, 1976; KALRA et al., 1975). Like other insect species, the lindane-resistant T. castaneum showed cross resistance to dieldrin (Bhatia & PRADHAN, 1972; BARWAL & KAIRA, 1982). However, there does not seem to be any information available on the mechanisms of dieldrin-resistance in this species. Hence, the present investigations were carried out to elucidate the role of metabolism and excretion of dieldrin as possible mechanisms of resistance in T. castaneum.

#### MATERIALS AND METHODS

14C-Dieldrin (85 mci/mmol) was obtained from Radiochemical Centre, Amersham (U K). Its purity was checked before use by thin layer chromatography system described below. Some of the non-radioactive metabolites of dieldrin namely, trans aldrindiol, 9-hydroxy dieldrin, photodieldrin and 4,5-aldrin-dicarboxylic dimethyl ester were supplied by the Institute of Ecological Chemistry, Munich, W. Germany. The X-ray plates for raidoautography were obtained from Hindustan Photo Film Manufacturing Co., India. Audits of Lindane resistant (R) and susceptible strain (S) of T. castaneum used in this study were from stock cultures maintained in this department; the detail regarding these were earlier given by BARWAL & Kalra (1982).

Metabolism: Batches of 100 aduts of both susceptible and resistant strains of T. castaneum were treated topically with  $1\mu$ 1 acetone solution of 14C-dieldrin at the dose of 0.1/1g per insect. The treated insects were held without food for 48 hours, after which those were homogenized along with their faeces in 10 ml of acetonitrile. The supernatant was separated after centrifugation of the homogenate. The residual material was rehomogenized twice with acetonitrile. The supernatant from the three spins were combined and then centrifuged at 1500 rpm for 10 minutes. The resultant supernatant was evaporated to near dryness under a stream of nitrogen. The residue was taken in 10 ml of petroleum ether and cleaned up by colum chromatography, using 'Florisil' as adsorbent (KLEIN et al., 1968). Elution was carried out first with 100 ml of 15 per cent ethyl ether and 0.3 per cent acetone in petroleum ether and then with 150 ml of acetone. These eluates were concentrated to small volumes with stream of nitrogen and then spotted on thin layer chromatoplates coated with silica gel G. The TLC plates were developed in the solvent system consisting of benezene-ethyl acetate (3:1,  $\nu/\nu$ ) and exposed to X-ray films for 15 days.

The silica gel areas corresponding to the spots on the X-ray film were separately scrapped and eluted in 10 ml acetone. The silica gel was removed by centrifugation and the supernatant was transferred to the scintillation vial. The vial was kept in an oven at 50°C to evaporate acetone. To this was added 10 ml of the scintillation cocktail prepared by dissolving 3 g of PPO and 100 mg of POPOP in 1 litre of toluene. This was shaken well and kept overnight at room temperature. The counts were recorded in Packard Liquid Scintillation Spectrometer Model 4430 in which the counts were corrected automatically for background.

The method of Singh & ThornHill (1980 a) was also used for the isolation and characterisation of metabolites of 14C-dieldrin formed by adults of the resistant strain. For this purpose, 550 adults of T. castaneum were treated topically with  $^{14}$ C-dieldrin at 0.1  $\mu_{\rm g/insect}$ At the end of 48 hours, the insects were ex. tracted. The purified extract was spotted on TLC plates and run using four different solvent systems, i.e. acetone-hexane (1:3, v/v), chloroform-ethatol (9:1, v/v), ether-hexane (1:1, v/v) and benzene-ethylacetate (3:1, v/v). The plates after development were exposed to X-ray films as described above for location of the spots. Authentic samples of metabolites of dieldrin were developed in similar solvent systems and detected by silver nitrate method (MATTHEWS & MATSUMURA, 1969).

#### Excretion:

Groups of 20 aduls of S- and R- strains were treated topically with 1  $\mu$ 1 acetone solution of  $^{14}$ C-dieldrin. Treated insects were kept in glass beakers and were not given any food. Groups of 20 insects were transferred to Potters' Elvejhem homogeniser at 4, 8, 24 and 48 hours after the treatment. These were

rinsed twice with 10 ml of acetone to remove the unabsorbed insecticide. The insects were then homogenised with 10 ml of acetone for 2 minutes and the homogenate was centrifuged at 1500 g for 2 mirutes. The residue was rehomogenised twice using 10 ml of acetone each time. The supernatants from all the three spins were then combined to give the internal (absrobed) insecticide. Faeces were collected from the glass beakers used to hold the insects with the help of camel hair brush and were then extracted following the method described for extraction of the absorbed insecticide. The holding beakers were rinsed with 10 ml of acetone to know the extent of removal of insecticide from insects as a result of abrasion. The extracts, thus oblained, were transferred to scintillation vials. The vials were kept in an oven at 50°C till the complete evaporation of solvoent. To each of the vial, 10 ml of scintillation cocktail was added. These were shaken well and kept overnight at room temperature before the counts were recorded.

#### RESULTS AND DISCUSSION

Adults of resistant strain of T. castaneum were found to form seven to eight metabolites of <sup>14</sup>C-dieldrin, of which two were identified as trans-aldrindiol and 9-hydroxy dieldrin (Fig. 1). Likewise Korte & Arent (1965) isolated six metabolites of dieldrin from the urine of rabbits after its oral administration. Nelson & Matsumura (1973) reported that Blattella germanica metabolised dieldrin into two mono-hydroxy dieldrins and trans-aldrindiol. Five metabolites of dieldrin were detected in Blaberus discoidalis (Singh & Thornhill, 1980 a), while seven were found in blowfly, Calliphora er vthrocephala (SINGH & THORNHILL, 1980 b).

The resistant strain of *T castaenum* was found to metabolize 22.5 per cent of the applied <sup>14</sup>C-dieldrin in 48 hours as compared to only 1.6 per cent by the susceptible strain (Table 1). Transaldrindiol was found to be the major metabolite in both the strains, but its amount

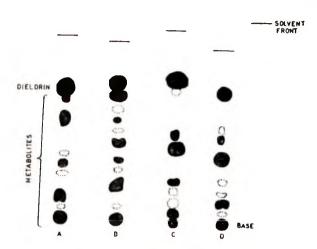


Fig. 1. Representatation of autoradiograms showing thin-layer chromatographic separation of 14c-dieldrin and its metabolites extracted from resistant *T. castaneum* using different solvent systems (A) acetone-hexane 1:3 v/v (B) chloroform-ethanol, 9:1 v/v (C) Etherhexane, 1:1 v/v (D) benzene-ethylacetate, 3:1 v/v. Soild spots- major metabolites; dotted spots- minor metabolites.

was significantly more in the R-strain than in the S-strain. Earlier, OONNITHAN & Miscus (1964) reported that <sup>14</sup>C-dieldrin was metabolized by dieldrin-resistant Culex pipiens quinquefasciatus to aldrindiol. On the other hand PERRY et al. (1964) reported taat 14C-dieldrin was neither metabolized nor excreted by any of the susceptible and dieldrin-resistant houseflies Later SELLERS & Musca domestica GUTHRIE (1972) reported the presence of polar metabolites of dieldrin in the resistant strains of houseflies at extended periods after treatment.

Dieldrin has been reported to stimulate synapses and sensory receptors and to induce the formation of multiple discharges after a long latency. However, transaldrindiol was found to exert this effect more quickly (WANG et al., 1971; VANDEN BERCKEN & NARAHASHI, 1974; NARAHASHI,

1976). It was, therefore, considered that trans-aldrindiol was an active form of dieldrin. According to Brooks (1980), the contribution of this metabolite to dieldrin action, however, remains conjectural. The hydroxylation of dieldrin to cis- and trans-aldrindiol has been reported to be mediated by the mixed-function oxidases (Nelson & Matsumura, 1973; MATTHEWS & Mc KINNEY, 1974). In view of the fact that the inhibitor of mixedfunction oxidases, sesamex exhibited small synergistic effect on dieldrin, the diol formation does not seem to be an activation reaction. Further, the amount of trans-aldrindiol found in the nerve cord as a result of in situ formation was too insufficient to account for toxicity since larger amounts of it entered the nervous system following topical application or injection without any poisoning effect (Brooks, 1977: Schroeder et al., 1977;

TABLE 1. Relative amounts of  $^{14}$ C dieldrin and its various metabolites in the resistant and susceptible strains of T. castaneum.

Per cent r	adioacti	ivity
resistant strain		susceptible strain
mean ± S D		mean ± S D
77.5* ± 2.1		$98.4 \pm 0.2$
$0.5 \pm 0.1$		$0.3 \pm 0.04$
$2.6 \pm 1.3$		$0.2 \pm 0.0$
$0.6 \pm 0.07$		ND
$15.1 \pm 0.7$		$0.6 \pm 0.07$
$1.1\pm0.3$		$0.3 \pm 0.04$
$1.7 \pm 0.6$		$0.2 \pm 0.03$
	resistant strain mean $\pm$ S D  77.5* $\pm$ 2.1  0.5 $\pm$ 0.1  2.6 $\pm$ 1.3  0.6 $\pm$ 0.07  15.1 $\pm$ 0.7  1.1 $\pm$ 0.3	mean $\pm$ S D  77.5* $\pm$ 2.1  0.5 $\pm$ 0.1  2.6 $\pm$ 1.3  0.6 $\pm$ 0.07  15.1 $\pm$ 0.7  1.1 $\pm$ 0.3

ND-not detected; SD-standard deviation: \* Man of two replications.

Table 2. Recovery of dieldrin (p moles) in external rinses, internal fraction and excreta of S- and R- strains of Tribolium castaneum over different time intervals.

Fraction	4 1	ır		3 hr		24 hr	4	8 hr
2.1	S	R	S	R	S	R	S	R
External	82.45*± 18.40	117.15± 6.28	50.71± 10.36	36 24± 3.86	23 45± 3.15	16 66± 3 33	26.09± 11.26	9.85±3.40
Internal	293.28± 41.09	137.51± 31.33	303,35± 12.69	162 49± 18.49	363.66± 49.45	73.60± 9.07	270.61± 56.67	33 78 <del>±</del> 16.54
Faeces	5.76± 1.41	18.73± 4.05	8 37± 0.68	37.21 ± 2.43	20.72± 4.66	135,81± 1.98	31.43± 8.91	182.09± 7.76
Container wash	8.24± 1.8	17,03± 0.57	8.43± 1.01	14.27± 1.37	11.70± 1.16	21.72± 1.83	16 02± 5.02	21 10± 1 42

<sup>\*</sup>Mean of three replications. ± standard deviation; S- Susceptible strain; R- Resistant strain.

SINGH, 1980). The high amount of trans-aldrindiol found in the R-strain of *T. castaneum* during the course of the present studies further support that the diol formation is a detoxication rather than an activation reaction.

The radioactivity found in the faeces of resistant strain was much higher than that in the faeces of the susceptible strain (Table 2), thereby suggesting the excretion of insecticide as a possible mechanism of dieldrin-resistance in *T. castaneum*. The

parent dieldrin was also detected in the excreta by gas-liquid chromatography. Earlier Rowlands et al. (1973) reported that the adults of resistant strain of *T. castaneum* excreted significantly higher amount of intact lindane than their susceptible counterparts. Resistant strains of houseflies have also been found to excrete more of <sup>14</sup>C-dield in than the susceptible strain (Winteringhan & Harrison, 1959; Sellers & Guthrie, 1972).

The internal level of <sup>14</sup>C-compounds in susceptible adults was more than in the resistant adults. However, this cannot be attributed to decreased penetration of the insecticide as the difference between the strains in the external/container wash was not much.

Acknowledgement: The investigations were in part financed by the Indian Council of Agricultural Research, New Delhi.

#### REFERENCES

- BARWAL, R. N. & R. L. KALRA (1982) Crossresistance characteristics of lindane-resistant and susceptible strains of *Tribolium castane*um (Herbst.) (Col: Tenebrionidae). *Entomon*, 7, 91—95.
- BHATIA, S. K. & S. PRADHAN (1972) Studies on resistance to insecticides in *Tribolium castaneum* (Herbst.). V. cross-resistance characteristics of a lindane-resistant strain. J. Stored Prod. Res., 8, 89—93.
- Brooks, G. T. (1977) Epoxide hydratase as a modifier of biotransformation and biological activity. *Gen. Pharmac.*, 8, 221—226.
- Brooks, G. T. (1980) Biochemical targets and insecticide action, 41-55, in: Insect Neurobiology and Pesticide Action (Neurotox '79) Soc. Chem. Ind. London, 489-496.
- CHAMP, B. R. & C. E. DYTE (1976) FAO global survey of pesticide susceptibility of stored grain pests. FAO Plant Prot. Bulk., 25, 1-49.
- KALRA, R., A. S. UDEAAN & O. S. BINDRA (1975) Status of insecticide resistance in

- stored grain insects in the Punjab. *Indian* J. Pl. Prot., 3, 186-196.
- KLEIN, A. K., J. D. LINK & N. F. IVES (1968) Isolation and purification of metabolites found in the urine of male rats fed with aldrin and dieldrin. J. Assoc. Off. Anal. Chem., 51 (4), 895—898.
- KORTE, F. & H. ARENT (1965) Metabolism of insecticides. IX (1) Isolation and identification of dieldrin metabolites from urine of rabbits after oral administration of <sup>14</sup>C-dieldrin. Llie Sci., 4 (21), 2017—2026.
- Matthews, H. B. & F. Matsumura (1969) Metabolic fate of dieldrin in the rat. J. agric. Fd Chem., 17, 845—852.
- MATTHEWS, H. B. & J. D. MCKINNEY (1974)
  Dieldrin metabolism to cisdihydroaldrindiol
  and epimerisation of cis-to-trans-dihydroaldrindiol by rat liver microsomes. *Drug Metab. Disp.*, 2, 333—340.
- NARAHASHI, T. (1976) Effect of insecticides on nervous conduction and synaptic transmission, 327—349. in: Insecticide Biochemistry and Physiology (Ed., C. F. WILKINSON) Plenum Press, Newyork and London.
- Nelson, J. D. & F. Matsumura (1973) Dieldrin (HEOD) metabolism in cockroaches and houseflies. Arch. envir. Contam. Toxicol., 1, 225—244.
- Oonnithan, E. S. & R. Miscus (1964) Metabolism of <sup>14</sup>C-dieldrin by dieldrin-resistant female adults of *Culex pipiens quinque-fasctatus*, J. econ. Ent., 57, 425-426.
- Perry, A. S., G. W. Pearce & A. J. Buckner (1964) The absorption, distribution and fate of <sup>14</sup>C-aldrin and <sup>14</sup>C-dieldrin by susceptible and resistant houseflies, *J. econ. Ent.*, 57, (6), 867—872.
- ROWLANDS, D. G., J. A. DALY & D. G. BLACKMAN (1973) Lindane resistance in the rust-red flour beetle. *Pest Infest. Control.*, 119, 1968—1970.
- Schroeder, M. E., D. L. Shankl & R. M. Hollingworth (1977) The effect of dieldrin and isomeric diols on synaptic transmission in the American cockroach and their relevance to dieldrin poisoning syndrom. *Pestic. Biochem. Physiol.*, 7, 403—415.

- Sellers, L. G. & F. E. Guthrie (1972) Distribution and metabolism of <sup>14</sup>C-dieldrin in resistant and susceptible housefly. *J.* econ. Ent., 65, 378—385.
- SINGH, G. J. P. (1980) Metabolism of <sup>14</sup>C-dieldrin in insect nervous system, 489-496, in: Insect Neurobiology and pesticids Action (Neurotox 79). Soc. Chem. Ind., London.
- SINGH, G. J. P & R. A. THORNHILL (1980 a) The uptake and break downg dieldrin in Blaberus discoidalis. Comp. Biochem. Physiol., 65, 47-52.
- Singh, G. J. P. & R. A. Thornhill (1980 b) Pharmacokinetics of dieldrin with special

- reference to its metabolism in the epicuticular wax layer in Calliphora erythrocephala. Gen. Pharmac., 11, 283—292.
- Vanden Bercken, J. & T. Narashashi (1974) Effects of aldrin-transdiol, a metabolite of tce insecticides dieldrin on nerve membrane. Eur. J. Pharmacol, 27, 255.
- WANG, C. M., T. NARAHASHI & M. YAMADA (1971) The neurotoxic action of dieldrin and its derivatives in the cockroach. *Pestic. Biochem. Physiol.*, 1, 84-91.
- WINTERINGHAM, F. P. W. & HARRISON (1959) Mechanism of resistance of adult houseflies to insecticides dieldrin. *Nature*, 184, 608—610.

## HOW MANY LARVAL INSTARS ARE THERE IN OPISINA ARENOSELLA?

#### P. B. SANTHOSH BABU & V. K. K. PRABHU

Department of Zoology, University of Kerala, Kariavattom, Trivandrum, India 695 581

(Received 7 November 1986)

Detailed laboratory studies at ambient conditions of 23—32°C and 75—90% RH (in Trivandrum, Kerala) at 12:12 photoperiod show that the insect O. arenosella has eight larval instars; the ratio of head capsule width of successive instars is more or less constant, ranging between 1.2 and 1.7; the graph showing log head capsule width and instar is a straight line.

(Key words: Opisina arenosella (Nephantis serinopa), coconut leaf eating (black headed) caterpillar, larval instars)

#### INTRODUCTION

Opisina arenosella Wlk. the black headed caterpillar of the coconut palm, has been reported to possess 5 larval instars (LEVER, 1969; NIRULA, 1956); six instars (NIRULA et al., 1951 as cited by RAMACHANDRAN et al, in their review, 1979) or even seven (ANTONY, 1962). However, more surprisingly, in our culture, we noted eight larval instars (unpublished observations). It is not uncommon for an insect species, especially belonging to Lepidoptera, to show variable number of instars in the life history. These supernumerary larval instars are induced by such factors as cooling (CYMBOROWSKI & BOGUS, 1976), chilling (PIPA, 1976), starvation (BHAS-KARAN & JONES, 1980; NIJHOUT, 1975) and injury (Krishnakumaran, 1972). Earlier workers on Opisina did not give any details of the method employed for their studies which were apparently rather casual laboratory observations. So it was thought that one or more of the above factors could be responsible for the discrepancy in the number of larval instars observed. It was found that this problem has not been carefully studied so far and so we conducted a detailed and careful study of the early life history (egg to pupa) of this insect in our laboratory and our findings are reported here.

#### MATERIALS AND METHODS

The stock culture of the insect was maintained in the laboratory (on natural food) at ambient temperature 23-31°C, 75-90% RH and 12:12 LD period approximately. The newly hatched 1st instar caterpillars were removed from this colony and their moult was followed and the number of instars counted. The observations were made for the duration of one year covering the peak period (from March to July) and lean period (from August to February). During this period of study, 7 batches each containing 8-10 larvae were examined, The larvae hatched from eggs were collected immediately and allowed to feed on 6 cm long pieces of matured coconut leaves. The leaves containing larvae were kept in clean specimen tubes (8 × 2 cm). Each specimen tube contained one larva. The specimen tubes were covered with white cloth, thus preventing escape of the larvae. The leaves were changed every day. The measurements were taken 10 h after ecdysis. The ecdysial day was designated as day-1. The width of head capsule was measured under a stereomicorscope with an ocular micrometer.

#### RESULTS AND DISCUSSION

Of the total of all observations, it was consistently found that in all individuals there were eight larval instars. The measurements of head casule width as well as length and breadth of the instar and its duration are shown in Table 1, which also indicate the ratio of the head capsule width of successive instars. Fig. 1

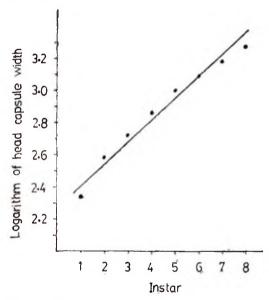


Fig. 1. Relationship between log head capsule width and instar in Opisina arenosella.

shows the relationship between larval stage and the log of head capsule width, which is almost a straight line, and the ratio of increase in head capsule width is 1.4 for most of the instars, except for the first to second instar, where it is 1.7 and during final three instars it is 1.2, 1.3 & 1.3. During the penultimate stages there is a tendency for decrease in the ratio from 1.4. On the whole, Dyers rule is found to hold good as shown in

Table and in Fig. 1. It is known that when there are supernumerary larvae due to insufficient nutrition, the ratio tends to be lower (SEAMENS & WOODRUFF, 1939). There are also other factors which could induce supernumerary instars as chilling (PIPA, 1976; NIJHOUT, 1975), (KRISHNAKUMARAN, 1972) and starvation (BHASKARAN & JONES, 1980). Though variable number of instars have been repored for this species by different workers viz; five (LEVER, 1969; NIRULA 1956), six (NIRULA et al., 1951 as reported RAMACHANDRAN et ul., 1979) or even seven (ANTONY, 1962), it my be noted that the above authors do not appear to have made any detailed studies on this problem and hence the discrepancy. For example, NIRULA et al. (1951) as reported by RAMACHANDRAN et al. (1979) recorded six larval instars for this insect, though according to NIRULA et al. themselves (Nirula et al., 1951) and Nirula (1956) it had only five larval instars and apparently RAMACHANDRAN et al. (1979) quoted NIRULA et al. (1951) erroneously. First instar of Nirula (1956) and Nirula et al. (1951) appears to consist actually of first and second instar of the present studies based on body length given by them (NIRULA has not given head capsule width); his second instar consists of the third and fourth instars; third instar corresponding to present fifth instar; fourth instar corresponding to sixth instar; his fifth instar corresponds to the present seventh and eighth instars. Apparently, Nirula's first, second and fifth instars are heterogeneous population made up of two larval instars each.

As mentioned previously many extraneous evironmental and nutritive factors affect the number of instars in the larval period. It may be noted that all the

т.	number	duration	head capsule	*RW	larval body	length (mm)
Instar ———	of insects used	(days)	width (mm)		lst day of instar	last day of instar
I	78	$3.33 \pm 0.47$	$0.22 \pm 0.02$		$1.54 \pm 0.10$	$2.69 \pm 0.24$
H	66	$3.50 \pm 0.50$	$0.38 \pm 0.03$	1.7	$2.77 \pm 0.30$	$3.78 \pm 0.38$
III	61	$3.77 \pm 0.46$	$0.52 \pm 0.02$	1.4	$3.93 \pm 0.39$	$5.24 \pm 0.37$
IV	56	$4.43 \pm 0.49$	$0.72 \pm 0.02$	1.4	$5.46 \pm 0.45$	$6.77 \pm 0.40$
V	52	$4.83 \pm 0.54$	$1.00 \pm 0.02$	1.4	$6.94 \pm 0.47$	$9.00 \pm 0.42$
VI	48	$5.44 \pm 0.50$	$1.22 \pm 0.02$	1.2	$9.11 \pm 0.46$	$12.02 \pm 0.43$

1.3

1.3

 $1.50 \pm 0.03$ 

 $1.87 \pm 0.05$ 

Table 1. Duration of larval instars, head capsule width and body length of Opisina arenosella larva on coconut leaves during development.

 $5.73 \pm 0.45$ 

 $7.57 \pm 0.75$ 

VII

VIII

44

above studies have been carried out in Our studies have been made throughout the year and we have not observed any individual variations with regard to the number of instars in this insect. So it appears reasonable that Opisina arenosella indeed possesses eight larval instars in its life history as evidenced by the present studies. None of the above factors appears to have affected the number of larval stages and this is supported by head capsule values and their ratio (WIGGLESWORTH, 1972). It is to be noted that the authors mentioned except Anrony (1962) have not stated the methods or materials they have employed ie., whether they are field observations or laboratory studies and if they are laboratory studies how they reared the larvae. Though the present method of rearing is uniform when compared to those of Antony (1962), who employed a number of methods for rearing the insect and the fairly large scale variation in his data is due apparently to the differing rearing conditions.

 $15.06 \pm 0.39$ 

 $18.95 \pm 0.47$ 

 $12.22 \pm 0.42$ 

 $15.28 \pm 0.42$ 

Acknowledgements: PBS is thankful to the Head of the Department for facilities and to CSIR for fellowship; VKKP acknowledges a Research Grant from DST. The paper was kindly gone through by Dr. G. B. PILLAI, Coconut Research Station, Kayamgulam.

#### REFERENCES

ANTONY, J (1962) Bionomics, Life history and Morphology of Nephantis serinopa M. a major pest. M. Sc. thesis, Kerala University, Trivandrum.

BHASKARAN, C. & G. Jones (1980) Neuroen-docrine regulation of corpus allatum activity in *Manduca sexta*. The endocrine basis for starvation induced supernumarary larval moult. J. Insect Physiol., 26, 431—440.

CYMBOROWSKI, B. & M. Bogus (1976) Juvenilizing effect of cooling on Galleria mellonella. J. Insect Physiol., 22, 669—672.

Krishnakumaran, A. (1972) Injury induced moulting in *Galleria mallonella* larvae. *Biol. Bull.*, 142, 281—292.

<sup>\*</sup> Ratio of head capsule width between successive instars.

- Lever, R. J. A. W. (1969) Pests of the Coconut palm. FAO agric, studies, 77, 66—69.
- Nuhout H. F. (1975) A threshold size for metamorphosis in the tobacco hornworm, Manduca sexta (L.). Biol. Bull., 149, 214— 225
- NIRULA, K. K. (1956) Investigation on the pests of coconut palm. 111. Nephantis serinopa Meyrick. Ind. Cocon. J., 9 (2), 101—131.
- NIRULA, K. K., J. ANTONY & K. P. V. MENON (1951) Investigation on the pests of the coconut palm. II. The coconut caterpillar Nepantis serinopa Meyrick. Ind. Cocon. J., 4(4), 217—223.
- PIPA, R. (1976) Supernumerary instars produced by chilled wax moth larvae: Endocrine mechanisms. J. Insect Physiol., 21, 1641—1647.
- RAMACHANDRAN, C. P., K. N. PONNAMMA, K. M. ABDULLA KOYA & C. KURIAN (1979) The coconut leaf-eating caterpillar, Nephantis serinopa Meyrick, a review. Phil. J. Cocon. Stud., 4(2), 9—17.
- SEAMANS, L. & L. C WOORDRUFF (1939) Some factors influencing the number of moults of the German Cockroach. J. Kansas ent. Soc. 12, 73-76.
- WIGGLESWORTH, V. B. (1972) The Principles of Insect Physiology (Seventh Edition). Chapman & Hall, Butler & Tanman, Frome.

#### **BRIEF COMMUNICATION**

## A NOTE ON THE OCCURRENCE OF ANOPHELES MINIMUS THEOBALD IN MANIPUR

K. B. RAJPUT<sup>1</sup> & T. K. SINGH

Department of Life Science, Manipur University, Canchipur, Imphal, Manipur, India 795 003

(Received 2 August 1985)

Collection record with its resting place of *Anopheles minimus* Theobald, 1901, an important malaria vector of North East Region was reported for the first time from Manipur. (Key words: record, *Anopheles minimus*)

Anopheles minimus Theobald, is one of the most dangerous malaria vectors in certain Southeast Asian countries and China. The species carries malaria in Burma, Formosa, South China, Indochina, Kampuchea, Vietnam and India. It plays an important role in transmitting malaria in Assam (Viswanathan et al., 1941), hilly tracts of Bengal (IYENGER, 1940), Tripura (MISHRA & DHAR, 1955), NEFA (MISHRA, 1950), Jeypore hilly tracts of Madhya Pradesh (Senior White, 1937; 1938) and in Singhbhum hilly region (SENIOR WHITE & DAS, 1938). The species has also been incriminated as malaria vector from Nagaland State (BHATNAGAR et al., 1982).

The anopheline records of Manipur State reviewed by Mortimer (1946) indicates the absence of suspected vectors of malaria of North East region i. e., A. balabacensis Baisas and A. minimus Theobald from records of earlier workers and from his own survey, for the State. Due to lack of any dissection record from the State there is no confirmed vector of

During recent years a survey work for mosquito fauna of Manipur was carried out and one female A. minimus Theobald specimen was collected from Moreh (150m MSL) on 19.viii.1984. The mosquito was collected from a shaded, slow moving nallah margin in dense shrubby forest area. Earlier reported domestic resting of the species from Assam (Christophers, 1933; Thomson, 1941) were later on doubted due to lack of sufficient information on outdoor and window-trap collections. The outdoor resting of the species have also been pointed out from Jeypore hilly tracts of Madhya Pradesh (Senior White, GHOSH & RAO, 1945) from China and variety flavirostris Ludlow, from Phillippines. From adjoining Burma, MACAN (1949), studied that the forested area

malaria. In view of the above circumstances A. minimus Theobald which is well known malaria vector in adjoining Burma and States of Assam & Nagaland, has always drawn keen attention of the earlier and present workers (MORTIMER Loc. cit.) but till present, due to its absence from faunistic records it was out of expectation to be vector for Manipur State.

<sup>1</sup> Present address: Regional Tasar Research Station, Manipur, India 795 002.

Kabaw valley it is outdoor rester but in open cultivated plains it is indoor rester. In order to organise any effective malaria control programme in the State it is of prime importance to confirm the vectorial role and the species behaviour in the State.

Acknowledgements: The authors are thankful for confirming the specimen to Dr. B. A. HARRISON, Manager, Walter Reed Biosystematics Unit, National Museum of Natural History, Washington D. C.

#### REFERENCES

- BHATNAGAR, V. N., S. R. DWIVEDI, D. G. MISRA & M. DAS (1981) Detection and incrimination of *Anopheles minimus* Theobald, 1901 as malaria vector in the foothill areas of Nagaland, India. *Ind. J. Mal.*, 19 (12), 129—134.
- COVEIL, G. (1944) Notes on the distribution, breeding places, adult habits and relation to malaria of the anopheline mosquitoes of India and the Far East. J. Mal. Inst. Ind., 5, 399-434.
- Christophers, S. R. (1933) The Fauna of British India including Ceylone and Burma, V. 4. Taylor and Francis, London, 360 pp.
- KRISHNASWAMY, A. K. (1961) Vectors of Malaria A. minimus Theobald, 1901., 91—98. In: Vectors of Malaria in India, Nat. Soc. Ind. Mal. Mosq. Dis., Delhi.
- IYENGAR, M. O. T. (1940) Further observations on Vectors of malaria in Bengal and notes

- on the seasonal infectivity of Anopheles. J. Mal. Inst. Ind., 3, 115-123.
- Senior White, R. (1937) On malaria transmission in Jeypore hills. Part I. A years' dissection result. Rec. Mal. Sur. Ind., 7, 47-75.
- SENIOR WHITE, R. (1938) On malaria transmission in Jeypore hills. Part II. A second years' result. J. Mal. Inst. Ind., 1, 129-145.
- SENIOR WHITE, R. & R. K. DAS (1928) On malaria transmission in Singhbhum Hills. J. Mal. Inst. Ind., 1, 169—184.
- SENIOR WHITE, R., A. R. GHOSH & V. V. RAO (1945) On the adult bionomics of some Indian anophelines with special reference to malaria control programme by pyrethrum spraying. J. Mal. Inst. Ind., 6, 129—215.
- MACAN, T. T. (1949) Malaria survey of the Arakon region of Bengal and Burma. *Parasitology*, **40**, 290—297.
- MISRA, B. G. (1956) Malaria in Northeast Frontier Agency (India). Ind. J. Mal., 10, 331-347.
- Mishra, B. G. & S. K. Dhar (1955) Malaria in Tripura State. *Ind. J. Mal.*, 9, 111-123.
- MORTIMER, D. A. (1946) Notes on the anopheline fauna of Manipur area, Eastern Assam. J. Mal. Inst. Ind., 6, 269-271.
- MUIRHEAD THOMSON, R. C. (1941) Studies on the behaviour of Anopheles minimus. Part V. The behaviour of adults in relation to feeding and resting in houses. J. Mal. Inst. Ind., 4, 217—245.
- RAO, T. RAMACHANDRA (1984) The Anophelines of India. (Rev. Ed.). Malaria Research Centre (ICMR), Delhi. 518 pp.

### OBSERVATIONS ON SPIDERS (ORDER: ARANEAE) PREDA-CIOUS ON THE COCONUT LEAF EATING CATERPILLAR OPISINA ARENOSELLA WLK. (NEPHANTIS SERINOPA MEYRICK) IN KERALA: FEEDING POTENTIAL\*

B. SATHIAMMA, S. P. JAYAPAL & G. B. PILLAI

Biological Control Laboratory, Central Plantation Crops Research Institute, Regional Station, Kayangulam, Krishnapuram, India 690 533

(Received 17 August 1985)

Twenty-six species of spiders belonging to twelve genera and six families were observed in association with the larval galleries of *Opisina arenosella* on coconut palms. Eighteen species were studied for their feeding potential under laboratory conditions. *Sparassus* sp., *Cheiracanthium* sp., *Rhene khandalensis* and *R. indicus* topped the list and *Neoscona elliptica* the last.

(Key words: Opisina arenosella, spider, coconut)

#### INTRODUCTION

A survey of the Opisina arenosella infested coconut gardens in Alleppey and Quilon Districts of Keraia revealed that twenty-six species of spiders belonging to twelve genera and six familes are frequently associated with the larval galleries of the pest (SATHIAMMA et al., unpublished). Some of them were observed to be highly predacious and they consume a huge population of the pest in the field. The objective of the present study was to evaluate the feeding potental of the spider predators associated with caterpillars of O. arenosella in the laboratory.

#### MATERIALS AND METHODS

Test spiders were collected individually from *Opisina* infested coconut plantations. They were reared separately in glass jar cages (size  $17 \times 6.5$  cm) in the laboratory. Coconut leaf-

let bits infested with known number of *Opisina* caterpillars were provided in each of the cages containing the spiders. The spiders were starved for a day prior to offering *Opisina* caterpillars to them. The number of caterpillars preyed upon and their feeding habits were recorded every day.

#### RESULTS AND DISCUSSION

All the eighteen spider species preyed on O. arenosella caterpillars (Table 1). The rate of predation varied from 0.05 to 1.54 caterpillars per day. Sparassus sp., Cheiracanthium sp., Rhene khandalensis and Rhene indicus recorded a high rate of predation, the rate being 1.54, 1.19, 0.71 and 0.70 caterpillars, respectively, per day. Neoscona elliptica consumed only 0.05 caterpillars per day and occupied the last rank in the rate of predation. interesting to observe that the ratio of predation varied considerably between sexes of the same species. Females of Sparassus sp., Cheiracanthium sp., Rhene indicus and Marpissa dhakuriensis consumed 1.54, 1.19, 0.70 and 0.62 caterpillars,

<sup>\*</sup> Contribution No.481, Central Plantation Crops Research Institute, Kasaragod - 670 124, Kerala, India.

TABLE 1. Feeding potential of spiders predacious on Opisina arenosella capterpillars in the laboratory (Mean of 10 replications).

SI. No.	Name of spider	Sex	Average no, of days observed	Average no. of prey con- sumed	No. of prey consumed per day
1	Sparassus sp.	F M	70 63	108	1.54
2	Cheiracanthium sp.	F M	57 66	72 68 46	1.14 1.19 0.70
3	Rhene khandalensis Tikader	F	38	27	0.71
4	R. indicus Tikader	F M	46 50	3 2 1 5	0.70 0.30
5	Plexippus paykulli (Aud.)	F	49	32	0.65
6	Marpissa dhakuriensis Tikader	F M	89 32	55 5	0.6½ 0.16
7	Tetragnatha andamanensis Tikader	F	65	38.5	0.59
8	Cheiracanthium melanostom Thorell	I	67	36	0.54
9	Clubiona drassodes Cambridge	F	28	12	0.43
10	Larinia jayasankari Biswas	F	78	31	0.40
11	Marpissa sp.	F	10	4	0.40
12	Phidippus bengalensis Tikader	F	48	18	0.36
13	Phidippus sp.	F	14	5	0.36
14	Phidippus sp.	F	9	3	0.33
15	Marpissa tigrina Tikdar	F	30	9	0.30
16	Marpissa sp.	F	10	3	0.30
17	Rhene danieli Tikadar	F	13	3	0.23
18	Neoscona elliptica Tikader Bal	F	20	1	0.05

F- female M- male I- immature stage

respectively, per day, whereas their male counterparts cosumed much less with 1.4, 0.7, 0.3 and 0.16 caterpillars per day. Rate of predation was observed to be very high when the prey caterpillar removed from the larval galleries were offered.

Spiders have been implicated as efficient biological control agents of pests of many agricultural crops. Howell & Pienkowski (1971) listed spider fauna of

Virginia alfalfa field and BAILEY & CADA (1968) on grain sorghum. Species of Cheiracanthium, Clubiona, Marpissa and Phidippus preyed on the maize borer larvae Chilo partellus and the consumption per predator varied from 2 to 18 borer larvae. Clubiona sp. and Marpissa spp. have been observed preying on the nymphs of Pyrilla perpusilla on sugarcane in Punjab (SINGH, 1967). JANDU (1972) observed Marpissa sp. and Phidippus sp. feeding on the

nymphs of Diaphorina citri on citrus. Phidippus audax (Hentz) consumed larvae of the boll weevil and salticid spiders on Heliothis spp. on cotton (WHITCOMB et al., 1963). Neoscona sp. preyed on aphids Myzus dycei, Chaitophorus kapuri, Liosomaphis himalayensis, and Metopolophium phaseoli on leguminous host plants including Urtica parviflora, Populus sp., Berberis sp. and Indet respectively, and Clubiona sp. on Aphis gossypii on Indet plant (Das & Chaudhuri, 1983). cona nautica Koch, R. Khandalensis Tikader and Thomisus sp. feed on aphids Macrosiphum rose and M. rosaeiformis on Rosa sp. (AGARWALA, 1983). A recent study Mohamed et al., 1982) of the natural enemies of the coconut caterpillar did not record any predatory spiders. The present report is the first record of the predacious spiders comprising major species such as Cheirncanthium, Rhene, Marpissa, Neoscona and Clubiona on the coconut leaf eating caterpillar Opisina arenosella.

The observations clearly revealed that the spiders constitute an important group of biocontrol agents exerting natural suppression on the field population of the coconut leaf eating caterpillar. Cheiracanthium, Sparassus and Rhene are the important predators consuming @ 0.7— 1.5 Opisina caierpillars per day. The magnitude of pest suppression these species can exert can further be seen from their long life span and their presence in the field almost throughout the year (SATHIAMMA et al., unpublished). During the period when Opisina population is very low, the spiders thrive on other species as well. Hence, conservation of these predators in the field is of great relevance. For the management of O. arensella, when chemical treatments are

to be done care should be taken to use only those chemicals which are less toxic to the spiders. The major handicap with the spider fauna is that they are not specific predators of the pest, but are polyphagous ones and a such they would feed on many species of other insects as well available in the field.

#### REFERENCES

- AGARWALA, B. K. (1983) Parasites and predators of rose infesting aphids (Homoptera: Aphididae) in India. *Entomon*, 8, 35-39.
- Bailey, C. L. & H. L. Chada (1968) Spider population in grain sorghum. Ann. ent. Soc. Am., 61, 567—571.
- Das, K. S. & D. R. CHAUDHURI (1983) Parasites and predators on aphids (Homoptera: Aphididae) from India—VI. New records of seven arachnids, one dipteran and one neuropteran predators from Himachal Pradesh, India. *Entomon*, 8, 27—34.
- HOWELL, J. O. & R. L. PIENKOWSKI (1971) Spider population in alfalfa, with notes on spider prey and effect of harvest. *J. econ. Ent.*, 63, 163—168.
- JANDU, M. K. (1972) Taxonomic studies on biological observation on the spiders predacious on citrus pests, M. Sc. Thesis, Punjab Agricultural University, Ludhiana.
- Mohamed, U. V. K., U. C. Abdurahiman & O. K. Remadevi (1982) Coconut caterpillar and its natural enemies. A study of the parasites and predators of Nephantis serinopa Meyrick. Zoological Monograph No. 2. 161 pp. University of Calicut, Kerala, India.
- SINGH, B., G. S. BATTU & A. S. ATWAL (1975) Studies on the spider predators of the maize borer *Chilo partellus* Swinhoe in the Punjab. *Indian J. Ent.*, 37, 72-76.
- Singh, G. (1967) Feeding behavior and sexual biology of sugarcae spider Clubiona drassodes Camb. (Clubinonidae: Araneae). M. Sc. Thesis, Punjab Agricultural University, Ludhiana.
- WHITCOMB, W. H., EXLINE & R. C. HUNGSIER (1963) Spiders of Arkansas cotton field. Ann. ent. Soc. Am., 56, 653—660.



## A NEW GENUS AND THREE NEW SPECIES OF ERIOPHYID MITES (ACARINA: ERIOPHYOIDEA) FROM WEST BENGAL INDIA

N. K. GHOSH & S. CHAKRABARTI

Biosystematics Research Unit, Department of Zoology, University of Kalyani, Kalyani, India 741 235

(Received 28 September 1985)

A new genus viz., Indosetacus under the subfamily Cecidophyinae and four new species of eriophyid mites viz., Indosetacus rhinacanthi, Acaphyllisa pipera and Rhombacus eucalypti are described from Purulia and Midnapore districts of West Bengal. Distribution, affinity and host-eriophyid relations of the new genus and species are also discussed. (Key words: Acarina, eriophyids, taxonomy, morphology, new genus, new species, India)

In this paper one new genus and three new species of eriophyid mites are described. The type slides are deposited presently in the collection of Biosystematics Research Unit, Department of Zoology, University of Kalyani, Kalyani, India 741 235.

#### Indosetacus gen. nov.

Body worm like. Rostrum and oral stylet short. Shield subtriangular, lacking anterior projection; dorsal tubercles at rear shield margin, setae directed caudad. Forecoxae contiguous, lacking an encircling line; sternal line short. Legs with all usual segments and setae, featherclaw simple. Thanosomal rings subequal dorsoventrally and completely microtuberculate but last few rings broader dorsally and not microtuberculated; ventral thanosome with all standard setae, female genitalia appressed to coxae; coverflap smooth: internal female apodeme short.

\* All measurements are in micrometers (\(m\)).

Type species: Indosetacus rhinacanthi sp. nov.

Remarks: Among the genera of Cecidophyinae, the present genus comes close to Cosetacus Keifer (1966), Paracolomerus Keifer (1975) and Circaces Keifer (1978) due to worm like body, shield without anterior projection, dorsal tubercles on rear shield margin and setae directed to rear. However, the new genus differs from Cosetacus by the presence of foretibial seta, sternal line and rear non-microtuberculated broad rings. It differs from Paracolomerus mainly by the absence of an encircling line on forecoxae and presence of rear non-microtuberculated broad rings and from Circaces by the presence of undivided body rings and third ventral setae.

#### 1. Indosetacus rhinacanthi sp. nov. (Fig. 1)

Female: Body 86—109\* long, 39-44 wide, worm like, whitish in colour. Rostrum 12—14 long, projecting forward; subapical seta short. Shield subtriangular without anterior projection, 21—23 long

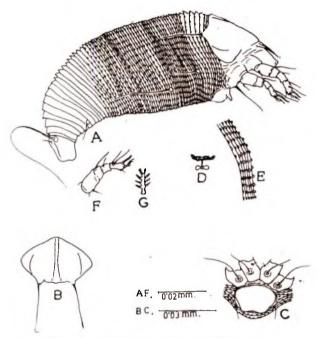


Fig. 1. Indosetacus rhinacanthi sp. nov.

Explanation of figures: A—Lateral view of mites; A<sub>1</sub>—Dorsal view of mites; B—Anterior dorsum of mites; C—Coxae with female genitalia; D—Internal female apodeme; E—Side skin structure; F—Fore leg; G—Featherclaw.

and 35-37 wide; shield design represents only a few lines; median line absent; admedians complete; submedians absent; lateral lines occur on rear 0.5 part of the shield; dorsal tubercles on rear shield margin; dorsal setae 16-25 long, directed caudad. Foreleg 16-24 long from trochanter base; femur 8-10 long, seta 9 long; patella 3.4—4.6 long, seta 9—12 long; tibia 2-4 long, seta short; tarsus 4-6 long, with two upper setae, each 11-14 long and one short lower seta; claw 6-7 long knobbed; featherclaw simple, 4-rayed. Hindleg 14—22 long from trochanter base; tarsus with an upper seta, 14-16 long and lower seta absent; other characters as in foreleg. Forecoxae joined by a short sternal line; all the three coxal setiferous tubercles outlining by the curve lines. Abdomen with about 65—74 microtuberculate rings except rear 10—12 rings which are broad and nonmicrotuberculate; microtubercles round and located within ring margin. Lateral seta 9—12 long, on about ring 11; first ventral seta 28—31 long, on about ring 28; second ventral seta 3—5 long, on about ring 44; third ventral seta 14—16 long, on about ring 6 from rear; caudal seta 51—58 long; accessory seta 5—7 long. Female genitalia 16—19 wide and 11—14 long, appressed to hind coxae; coverflap smooth; genital seta 4—6 long. Internal female apodeme shortened in ventral view.

Male: Unknown.

Holotype: Q, on slide (No. 710/1/85), INDIA: WEST BENGAL: Purulia, Manbazar, 15.i.1985 ex Rhinacanthus nasuta, (Acanthaceae), coll. N. K. Ghosh.

Paratypes: Many QQ, on the holotypic slide and on 5 slides (Nos. 711—715/1/1985), collection data as in the holotype. *Distribution*: India: West Bengal.

Relation to the host plant: The mites make pouch galls on both surfaces of leaves. Cavity of galls are covered with white hairy outgrowths. During heay infestation, the entire leaf surface becomes twisted and distorted.

#### 2. Acaphyllisa pipera sp. nov. (Fig. 2)

Female: Body 142—155 long, 40-46 wide, fusiform, brownish in colour. Rostrum 16—19 long, curved down; subapical seta 5 long. Shield trianguler with proxi-

mally pointed anterior lobe over gnathosome base, 35-42 long and 42-44 wide; shield design represents a few lines and granules; median line present indistinctly on anterior 0.25 part of the shield; admedians complete; submedians complete and directed to the lateral shield; shield granulated laterally dorsal tubercles ahead of rear shield margin, setae 4-6 long, directed up and centrad. Foreleg 24-30 long from trochanter base; femur 9-12 long, seta 7—9 long; patella 3—5 long, seta 21-25 long; tibia 5-6 long, seta 2-4 long; tarsus 5 long with 2 upper setae, 15-21 long and a short lower seta; claw 3-5 long, knobbed; featherclaw divided and 4-rayed on each side. Hind leg 22-26 long from trochanter base with usual segments and setae.

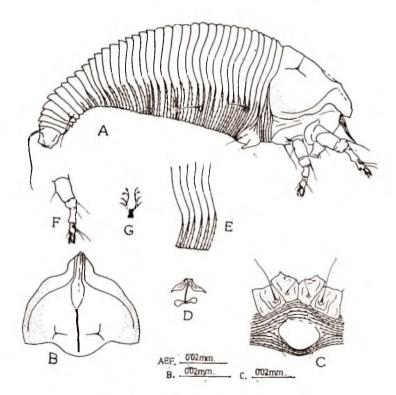


Fig. 2. Acaphyllisa pipera sp. nov. For explanation of figures, see Fg. 1.

Forecoxae connate, sternal line distinct; both the coxae ornamented with lines and granules; first setiferous coxal tubercles at the level of anterior sternal end; second coxal tubercles a little ahead of the transverse line across third coxal tubercles.

Abdomen with about 35 smooth tergites and 72 microtuberculate sternites; microtubercles are rounded and present on ring margin; thanosome with a middorsal longitudinal ridge which is fading gradually to rear. Lateral seta 8—9 long, on about sternite 10; first ventral seta 14—18 long, on about sternites 26; second ventral seta 7—9 long, on about sternite 42; third ventral seta 14–18 long, on about

5 from rear, accessory seta absent. Female genitalia 11—14 long, 16—18 wide; coverflap smooth; internal female apodeme moderate; genital seta 8—10 long.

Male: Unknown.

Holotype: Q, on slide (No. 752/10/85), INDIA: WEST BENGAL: Purulia, Salpara, 1.ii.1985 ex *Piper betle* Linn., (Piperaceae), coll. N. K. Ghosh.

Paratypes: Many QQ, on the holotypic slide and on 5 slides (Nos. 753—757/10/85), collection data as in the holotype.

Distribution: India: West Bengal.

Relation to the host plant: The mites the ventral surface of leaves as simple leaf vagrants. Due to infestation, leaves turn brownish.

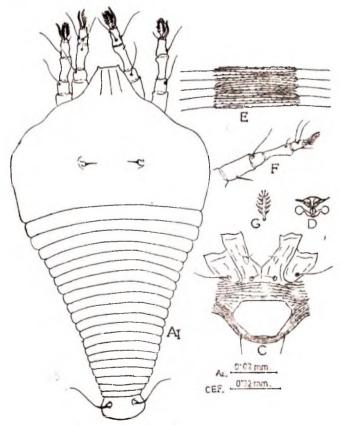


Fig. 3. Rhombacus eucalypti sp. nov. For explanation of figures, See Fg. 1.

Remarks: This species is closely related Acaphyllisa distasa Keifer (1961) and A. parindiae Keifer (1978) by the presence of first setiferous coxal tubercles, granulated lateral shield and coxae. But differs from both the above species by the presence of proximally pointed shield lobe, number of rays in featherclaw and smooth genital coverflap. In addition, it also differs from distasa by smooth tergites and absence of accessory seta and from Parindiae by round microtubercles.

#### 3. Rhombacus eucalypti sp. nov. (Fig. 3)

Female: Body 139—146 long, 70— 74 wide, robust, flattened, rhombic form, brownish in colour. Rostrum 21—25 long. curved down; subapical seta short. Shield subtriangular, 53-56 long, 74-83 wide, with a broad anterior lobe; shield surface more or less smooth; median and admedians occur only in lobe area; lobe in dorsal view with 2 indentations; dorsal tubercles well ahead of rear margin, setae 5 long, directed up. Foreleg 42-44 long from trochanter base; femur 11-13 long, seta 16 long; patella 4-6 long, seta 16 long; tibia 10 long, seta short; tarsus 8 long and with 2 upper setae, each 16-19 long and a short lower seta; claw 7 long, simple; featherclaw simple, 6-rayed. Hind leg 37-39 long from trochanter base; tarsus 7 long with one upper seta, 18 long and a small lower seta; claw 8 long, simple; other characters as in fore leg. Forecoxae connate, sternal line short; first coxal setiferous tubercles set ahead of sternal end level; second coxal tubercles at the level of third coxal tubercles, second coxal setae very small.

Abdomen with about 18—22 broad, smooth tergites and 60—70 microtuber-culate sternites; microtubercles pointed

and located within ring margin. Lateral seta 19—21 long, on about sternite 5; first ventral seta 21—26 long, on about sternite 20; second ventral seta 19 long, on about sternite 39; third ventral seta 28—30 long, on about sternite 6 from rear; accessory seta absent. Female genitalia 20—23 wide and 12—14 long; genital coverflap smooth; internal female apodeme large; genital seta 12 long.

Male: Unknown.

Holotype: Q, on slide (No. 793/19/85), INDIA: WEST BENGAL: Midnapore, Hoomgarh, 27.ii.1985 ex Eucalyptus globulus Labill., (Myrtaceae), coll. N. K. Ghosh.

Paratypes: Many QQ, on the holotypic slide and on 4 slides (Nos. 794—797/19/85), collection data as in holotyye.

Distribution: India: West Bengal.

Relation to the host plant: This species is a leaf vagrant and inhabits on ventral surface of leaves. No remarkable damage symptom was observed though the population was heavy.

Remarks: So far, 3 species, viz., Rhombacus morrisi Keifer (1965), R. asclepiadii Keifer (1969) and R. rheumella Keifer (1970) are known under the genus Rhombacus Keifer (1965). The present species differs from all the above species in having 6-rayed featherclaw, smooth genital coverflap, short second coxal seta and by the shield design.

Acknowledgements: The authors express thanks to the University Grants Commission, New Delhi, for financing the work and to the Head, Department of Zoology, University of Kalyani, for Laboratory facilities.

#### REFERENCES

Keifer, H. H. (1961) Eriophyid studies B-4. Bur. Ent. Calif. Dept. Agric., 1-20.

Keifer, H. H. (1965) Eriophyid studies B-14. Bur. Ent. Calif. Dept. Agric., 1—20.

Keifer, H. H. (1966) Eriophyid studies B-20. Bur. Ent. Calif. Dept. Agric., 1-20.

Keifer,, H. H. (1969) Eriophyid studies C-2 Agric. Res. Serv. U.S. Dept. Agric., 1-20.

Keifer, H. H. (1970) Eriophyid studies C-4.

Agric. Res. Serv. U.S Dept. Agric., 1-24.

Keifer, H. H. (1975) Eriophyid studies C-10. Agric. Res. Serv. U.S. Dept. Agric., 1-24.

Keifer, H. H. (1978) Eriophyid studies C-15. Agric, Res. Serv. U.S. Dept. Agric., 1-24.

# EFFECT OF MATING DURATION ON FECUNDITY AND FERTILITY OF EGGS IN BOMBYX MORI L (LEPIDOPTERA: BOMBYCIDAE)\*

M. THOMAS PUNITHAM, M. A. HANIFFA & S. ARUNACHALAM

Post Graduate & Research Department of Zoology, St. Xaviers' College,

Palayamcottai, India 627 002

(Received 17 August 1985)

Virgin Bombyx mori took 21 h to lay 176 eggs per individual and the percentage of hatching was nil. Mating duration upto 6 h reduced the preoviposition period and increased the total egg output by three times but subsequent increase in duration resulted in negative effects; it produced enhanced values in hatching upto three h duration. Increase in mating duration caused a decrease in body weight of females. Post oviposition life span was maximum (261 h) for virgin B. mori and decreased as a function of mating duration. Body weight of first instar progeny increased from 2.96 mg to 3.31 mg live weight with an increase in mating duration from 3 to 12 hrs.

(Key words: mating, preoviposition period, eggs, hatching)

#### INTRODUCTION

Mating in insects provides a stimulus for oogenesis (BENR, 1969; PICKFORD et al., 1968) and oviposition (LEAHY & Lowe, 1967). GILLOT & FRIEDEL (1971) reported a linear relationship between number of matings and egg output (DAVEY. 1967; GORDON & BANDAL, 1967). Only a few reports are available on the effects of consecutive matings (SUBRA MANIUM et al., 1980) and mating duration (SHAHI & KRISHNA, 1979) on total egg output and hatching in insects. But these reports do not reveal any information about the weight and life span of the mated female and/or the newly hatched young ones. The present investigation reports egg production, hatching, weight of newly hatched instars and weight and life span of female silk worm Bombyx mori as a function of mating duration.

#### MATERIALS AND METHODS

Healthy, fresh cocoons of B. mori of weight 1800 ± 100 mg were purchased from the State Sericulture Farm (Nannaharm, Tirunelveli, Tamil Nadu) and acclimated to Laboratory conditions. After emergence males and females were separated and the females were segregated into 13 groups, each containing 3 individuals. Test individuals of the first group (virgin) were allowed to lay eggs without mating. The females of the second and third groups were allowed to mate for 5 and 10 minutes respectively. Similarly for the succeeding groups, mating duration was increased by 10 minutes upto 1 hr, 120 minutes 3 h and 180 minutes upto 30 h respectively. After mating the male B. mori. was not allowed to mate another female and was discarded immediateely. Mated females were placed separately on rough brown sheets and were allowed to lay eggs by placing inverted plastic containers over them. The time taken by the imago after emergence till it started egg laying (preoviposition period) was recorded for each female. The number of eggs laid was counted and the percentage of hatching was calculated. After oviposition each female was

TABLE 1. Effect of mating duration on preoviposition period, egg output, hatching, postoviposition weight and life span and body weight of first instar in B. mori.

Mating time (min)	Preoviposition period (h)	Number of eggs laid	Percentage of hatch- ing	Weight after oviposition (mg live wt)	Post-ovipo- sition life span (h)	Body wieght of first instar (mg live wt)
0	21.4 ± 4.90	175.7 ± 12.47	0	736.3 ± 18.63	261 ± 29	
5	$12.8 \pm 2.60$	$143.3 \pm 14.09$	0	716.6 ± 16.27	$237 \pm 14$	
10	$11.8 \pm 0.44$	$167.0 \pm 11.77$	0	$709.5 \pm 18.35$	235 ± 5	
20	9.1 ± 0 61	$202.7 \pm 10.70$	0	669.4 ± 18.10	198 ± 4	
30	$9.0 \pm 0.57$	$301.0 \pm 14.37$	0	$554.5 \pm 15.78$	171 ± 28	
40	$8.9 \pm 0.49$	$342.1 \pm 17.25$	0	414.3 ± 19.24	145 ± 17	
50	$8.7 \pm 0.58$	$388.0 \pm 13.29$	0	$332.7 \pm 18.40$	149 ± 4	
60	$7.7 \pm 0.62$	$407.3 \pm 13.74$	0	296.3 ± 11.34	$131 \pm 12$	
180	$7.9 \pm 0.44$	$539.2 \pm 23.50$	82.5 ± 8.4	228.9 ± 11.17	128 ± 1	$2.96 \pm 0.14$
360	$7.9 \pm 0.63$	$546.0 \pm 25.50$	$88.6 \pm 7.3$	219.8 ± 12.92	124 ± 4	$2.93 \pm 0.13$
540	$9.2 \pm 0.81$	$532.2 \pm 9.50$	$96.7 \pm 1.3$	220.6 ± 14.72	129 ± 9	$2.91 \pm 0.19$
720	$12.4 \pm 0.46$	$529.3 \pm 10.21$	$81.6 \pm 12.2$	$224.9 \pm 15.81$	151 ± 4	$2.95 \pm 0.18$
900	$15.2 \pm 0.67$	487.1 ± 17.84	$91.2 \pm 6.9$	$262.9 \pm 16.94$	140 ± 12	$3.07 \pm 0.17$
1080	$18.9 \pm 0.12$	$462.3 \pm 28.53$	$84.5 \pm 7.3$	$320.0 \pm 15.82$	174 ± 12	$3.14 \pm 0.16$
1260	$21.3 \pm 0.07$	445.4 ± 19.26	77.8 ± 7.5	325.9 ± 12.32	156 ± 17	$3.20 \pm 0.14$
1440	$24.4 \pm 0.06$	$433.7 \pm 21.50$	$64.4 \pm 8.0$	$335.5 \pm 6.88$	126 ± 19	$3.29 \pm 0.13$
1620	$27.8 \pm 0.21$	$445.3 \pm 17.70$	$66.1 \pm 10.6$	$288.4 \pm 23.72$	$168 \pm 10$	$3.29 \pm 0.18$
1800	$30.2 \pm 0.43$	$380.1 \pm 30.21$	65.9 ± 6.1	221.2 ± 16.34	148 ± 17	$3.31 \pm 0.12$
	p < 0.001	<i>p</i> < <b>0</b> .001	p < 0.001	p < 0.05	p > 0.05	p > 0.05

Values represent the mean  $\pm$  SD for the number of observations on the test individuals.

weighed in a monopan balance (sensitivity: 0.01 mg) and life span of the female was recorded separately.

#### RESULTS AND DISCUSSION

Virgin B. mori took 21 h to lay eggs after the emergence. Increase in mating duration upto 6 h reduced the preoviposition period and after that the time increased. Virgin B. mori laid 176 eggs/individual and the percentage of

hatching was nil. The female mated for 1 h took the minimum time of 7.7 h to lay eggs. Mating duration increased the total egg output by 3 times upto 6 h (546 eggs/female) and subsequent increase in mating duration upto 30 h failed to produce any further significant increase in total egg output. Mating is not obligatory for either oviposition or egg production in *B. mori*. The complete failure

of hatching of eggs laid by the virgins shows clearly that insemination is essential for further development. Increase in mating duration produced enhanced values in hatching from 83% in females mated for 3 h to 97% in those mated for 9 h and thereafter subsequent increase in mating time caused a decrease in the percentage of hatching (Table 1).

Virgin B. mori showed the maximum live weight of 736 mg. Increase in mating duration caused a decrease in body weight of female to 220 mg when mated for 6 h to 30 h with slight fluctuations in between. Post-oviposition life span was

maximum (261 h) for virgin *B. mori* and it decreased to 50 percent when mated for 1 hr and after that increase in mating duration produced elevated and reduced values in life span. First instar progeny of females mated for 3 to 12 h did not show much variation in their body weight. Maximum body weight of 3.31 mg live weight was noticed for the first instar larvae belonging to females mated for a maximum duration of 30 h.

Increase in egg production observed in mated *B. mori* as a function of mating duration supports the previous observations. For instance the number of eggs

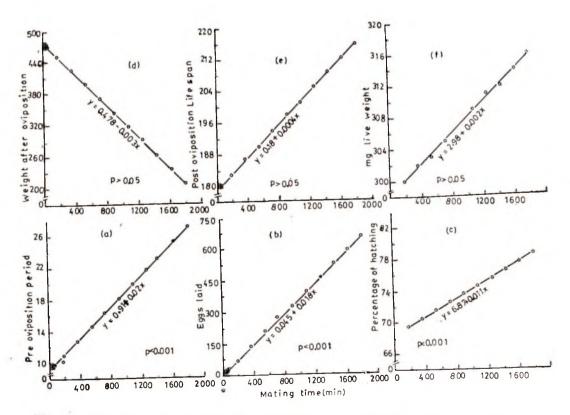


Fig. 1. Regressions and student's 't' as a function of mating time versus pre-oviposition period (a), egg output (b), hatching (c), post oviposition weight (d), life span (e) and live weight of first instar (f) in B. mori.

deposited by *Dysdercus koeinigii* increased from 14 to 218 per female with the increase in mating duration from 2 to 10 h (Shahi & Krishna, 1979). The increase in egg production observed in mated female insects could be due to the fecundity enhancing substance of accessory secretion of male insects (GILLOT & FRIEDEL, 1977) and/or due to activation of corpus allatum (Davis, 1965).

Increase in mating duration from 3 h to 15 h produced elevated values in percentage of hatching and after 15 h, subsequent increase in mating duration caused negative effects such as decrease in egg output, hatching, post oviposition life span and post oviposited female body weight and increase in preoviposition period (SUBRAMANIUM et al., 1980). The mated females lost considerable body weight (about 60%) and life span (about 50%) after oviposition and this loss appears to be relatively more when fertile eggs are laid. Since B. mori does not feed in the imaginal stage, it is possible to suggest that the decrease in body weight and life span as a function of mating duration could be due to energy expenditure and exhaustion. Regressions plotted (Fig. 1) confirmed that mating duration influenced preoviposition period (p < 0.001), egg production (p < 0.001), percentage of hatching (p < 0.001), post oviposition weight (p > 0.05) and life span (p > 0.05) significantly.

Acknowledgements: We are grateful to Prof. T. J. PANDIAN (Madurai Kamaraj University) and Prof. P. S. RAMAMOORTHY (Hyderabad) for valuable guidance, helpful suggestions and support. Thanks are due to Prof. R. K. RAMKUMAR (Fisheries College, Tutiucorin) for statis-

tical analyses and Mr. Murugesan (SRF, CSIR) for help.

#### REFERENCES

- BENZ, G. (1969) Influence of mating, insemination and other factors on oogenesis and oviposition in the moth Zeiraphera diniana.

  J. Insect Physiol., 15, 55-71.
- Davey, K. G. (1967) Some consequences of copulation in *Rhodnius prolixus*. J. Insect Physiol., 13, 1629—1636.
- Davis, N. T. (1965) Studies of the reproductive physiology of cimicidae (Hemiptera). II. Artificial insemination and the function of tne seminal fluid. J. Insect Physiol., 11, 355—366.
- GILLOT, C. & T. FRIEDEL (1977) Fecundity enhancing and receptivity inhibiting substances produced by male insects, 199-218, in: Advances in Invertebrate Reproduction (ed. K. G. ADIYODI & R. G. ADIYODI) Vol. I, 199-218 pp.
- Gordon, H. T. & S. K. Bandal (1967) Effects of mating on egg production by the large milk weed bug *Oncopeltus fasciatus* (Hemiptera: Lygaeidae). *J. econ. Ent.*, 60, 1099—1102.
- LEAHY, M. G. & M. L. LOWE (1967) Purification of the male factor increasing oviposition in *Drosophila melanogaster*. *Life*, *Sci.*, 6, 151-156.
- PICKFORD, R., B. EWEN A. L. & C. GILLOT (1969) Male accessory gland substance: an egg laying stimulant in *Melanoplus sanguine-pes* (F) (Orthoptera: Acrididae). Can. J. Zool., 47, 1199—1203.
- PRATT, G. E. & K. G. DAVEY (1972) Cropus allatum and oogenesis in *Rhodnius prolixus* (Stal). 3. The effect of mating. *J. exp. Biol.*, 56, 223—237.
- SHAHI. K. P. & S. S. KRISHNA (1979) Oviposition and fertility of eggs in *Dysdercus koenigii* (Fabr). *Mitt. Zool. Mus. Berlin.*, 55, 291—296.
- TAYLOR, O. R. Jr. (1967) Relationship of multiple mating to fertility in Atteva punctella (Lepidoptera: Yponomeutoidae). Ann. ent. Soc. Am., 60, 583—590.

# STUDIES ON HOST AGE PREFERENCE AND BIOLOGY OF EXOTIC PARASITE, COTESIA MARGINIVENTRIS (CRESSON) (HYMENOPTERA: BRACONIDAE)\*

S. K. JALALI, S. P. SINGH & CHANDISH R. BALLAL

All India co-ordinated Research project on Biological Control of Crop Pests and Weeds, Indian Institute of Horticultural Research,
Hessaraghatta Lake Post, Bangalore, India - 560 089

(Received 28 September 1985)

Cotesia (=Apanteles) marginiventris (Cresson), a solitary endo-parasite was introduced in India for trials against tobacco caterpillar, Spodoptera litura (F.). In an effort to evaluate mass rearing technique, studies on host age preference and biology of this parasite were conducted in laboratory where the temperature was  $27\pm1^{\circ}\text{C}$  and RH 50-60 per cent. For oviposition, 3 to 5 days old larvae were preferred. Mean developmental period from oviposition to cocoon formation lasted  $8.2\pm1$  days and pupal period 3 to 4 days. Sex ratio was 1:0.25 (male:female). A single mated female was capable of parasitising on an average 91.3 larvae during its life span. Mean longevity of adult female and male was 5.3  $\pm$  0.81 and 4.0 $\pm$ 0.89 days respectively.

(Key words: host age preference, biology, Cotesia marginiventris, Spodoptera litura).

#### INTRODUCTION

Spodoptera litura (F.) (Lepidoptera: Noctuidae) is a serious polyphagous pest. It has been reported feeding on 112 species of plants belonging to 44 families (Moussa et al., 1960). Indigenous natural enemies are not successful in satisfactorily suppressing this pest. Therefore, under All India Co-ordinated Research Project on Biological Control of Crop Pests and Weeds, C. marginiventris (MASON, 1981 has redefined the subfamily Microgastrinae and revived among others, genus Cotesia Cameron and placed the species marginiventris Cresson creating a new combination - C. marginiventris (Cress.); native of West Indies (MUESEBECK, 1921),

was imported for trials against S. litura in 1981. Though, it was earlier introduced in India in 1969 (RAO et al., 1971), no efforts were made to multiply it on a large scale for field evaluation. C. marginiventris is a major larval parasite of green clover worm, Plathypena scabra (F.) (MUELLER & KUNNALACA, 1979) in Arkansas, U. S. A. and S. Carolina (Mc-CUTCHEON & TURNIPSEED, 1981). BOLING & PITRE (1970) have studied life history and immature stages of this parasite on Pseudoplusia includens (Walker), Trichoplusia ni and Heliothis virescens (F.) and KUNNALACA & MUELLER (1979 on Plathypena scabra (F.). However, in India, information on host age preference and biology of C. marginiventris was not Therefore, these laboratory available. studies were conducted utilising tobacco caterpillar as a host.

Contribution No. 120/85 of the Indian Institue of Horticultural Research, Bangalore.

#### MATERIALS AND METHODS

Host insect, S litura was multiplied in the laboratory (average temperature  $27\pm1^{\circ}\text{C}$  & R H 50-60%) on an artificial diet (based on chick pea flour) developed for Heliothis armigera (Hb) by NAGARKATTI and SATYAPRAKASH (1974). Parasites used in the present study were obtained from laboratory where these are now continuously reared on S. litura larvae. Freshly emerged males and females of C. marginiventris in the ratio of 1:1 were held for 24 hours in glass vials (15 × 2.5 cm) for mating. Fifty per cent honey + water solution was provided on waxed paper strips inside the vials as adult food.

#### Host age preference:

Study was conducted by exposing one to ten days old S. litura larvae to C. marginiventris for 24 hours. 100 S. litura larvae of each age were released separately on castor leaves kept inside transparent plastic containers (20×16 cm). In each container 4 (one day old) mated females of C. marginiventris were inintroduced. Each treatment was replicated 5 times. After exposure, parasitised S. litura larvae were collected and reared individually on artificial diet in glass vials (7.5 × 2.5 cm). Each larva was observed daily for cocoon formation and subsequently cocoons were observed for adult emergence.

#### Biology

This study was conducted to determine developmental period (from oviposition to cocoon formation). Three day old larvae of S. litura were exposed individually to mated females of C. marginiventris. S. litura larvae were held with a camel brush and were exposed to mated females of C. marginiventris individually. Larvae were exposed to the parasite thrice a day-at 9.00 AM, 12 noon and 3 PM. Parasitised larvae were transferred on artificial diet in glass vials (7.5×2.5 cm). Larvae were observed for cocoon formation and subsequently cocoons were observed for adult emergence.

#### Fecundity and longevity:

This experiment was conducted by employing two methods. In the first method, 3 day old larvae were exposed to single mated female of *C. marginiventris* as described above.

exposed till female parasite Larvae were stopped pricking. This was repeated daily till female died. This treatment was replicated six times. In the second method, fifty 3 day old S. litura larvae were released on foliage of castor leaves in transparent plastic container (20×16cm) to which a single mated female of C. marginiventris was introduced for 24 hours. This exposure was repeated daily till female died. This experiment was also replicated six times. After exposure to the female parasite, all the larvae were reared individually on artificial diet in glass vials (7.5×2.5 cm). Larvae were examined daily for cocoon formation.

#### RESULTS AND DISCUSSION

#### Host age preference:

C. marginiventris preferred 3 to 5 day old larvae of S. litura. Parasitism obtained for 3, 4 and 5 day old larvae was 67.2, 52.0 and 43.0 per cent respectively (Table 1). Percentage parasitism reached its peak on 3 day old larvae and after that there was gradual decline upto 10th day. Eight to 10 day old larvae were less preferred as compared to 1 day and 2 day old larvae. KUNNALACA & MUELLER (1979) reported that the 1st instar larvae of P. scabra were most preferred. BOLING & PITRE (1970) reported preference to 2 days old larvae of T. ni. Kunnalaca & Mueller (1979) had also reported the increase of parasitism from 27.8 to 88.9 percent when exposed for one and 24 hours reseectively. Sex ratio obtained has also been furnished in Table 1. In general, higher percentage of females were obtained when 3 to 6 day old larvae were parasitised.

#### Developmental period:

Mean developmental period obtained from the time of oviposition to cocoon formation was  $8.2 \pm 1$  days with more number of parastic larvae emerging on 8th day. Pupal stage lasted 3 to 4 days

_	Age of the	Total number	100	Sex-rat	io (	of adults
Exposure time(h)	host larvae in days	of host larvae parasitised out of 500 larvae	parasitism	male	:	female
24 hours	1	23	4.6	1	:	.06
	2	98	19.6	1	:	.11
	3	336	67.2	1	:	.24
	4	260	52.0	1	:	.27
	5	210	42.0	1	:	.26
	6	138	27.6	1	:	.24
	7	66	13.2	1	:	.24
	8	10	2.0	1	:	.14
	9	7	1.4	1	:	0
	10	3	0.6	i	:	0

TABLE 1. Host age preference of C. marginiventris.

with 70 per cent adult emergence on 3rd day. Boling & Pitre (1970) reported mean development time from oviposition to cocoon formation in case of *H. virescens* as 6 days and 7 days each in case of *T. ni.* and *P. includens*. Kunnalaca & Mueller (1979) reported mean developmental time from oviposition to parasite larval emergence as 8 days at 30°C and 11 days at 25°C and pupal period as 3-5 days at 30°C and 4—7 days at 25°C in case of *P. scabra*.

#### Fecundity and longevity:

In the first treatment where larvae were exposed individually,  $111.8 \pm 16.8$  (91 to 139) larvae were parasitised. In second treatment where larvae exposed on mass  $70.8 \pm 10.4$  (59 to 87) larvae were parasitised which was quite low as compared to individual exposure method. The longevity of adult female and male was  $5.3 \pm 0.81$  and  $4.0 \pm 0.89$  days respectively in presence of the host. Anonymous (1984) reported that the adults lived for 15-18

days in presence of the host. However, in the present study not even a single female lived for such a long time. KUNNALACA & MUELLER (1979) observed the mean longevity at 30°C and 25°C as 5.6 nnd 9 days respectively.

Acknowledgements: The authors are grateful to Dr. F. D. BENNETT, Director, Commonwealth Institute of Biological Control for arranging the shipment of the parasite and to Dr. K. L. CHADHA, Director, Indian Institute of Horticultural Research, Bangalore for providing facilities. The assistance provided by Mr. M. B. BIRADAR, Senior Techical Assistant (T-4) is also gratefully acknowledged.

#### REFERENCES

Anonymous (1984) Annual Report of All India Co-ordinated Research Project on Biological Control of Crop Pests and Weeeds, II HR., Bangalore.

Boling, J. C. & H. N. PITRE (1970) Life-history of Apanteles marginiventris (Cresson) with description of immature stages. J. Kans. Ent. Soc., 43, 465—470.

KUNNALACA, S. & A. J. MUELLER (1979) A laboratory study of Apanteles marginiventris,

- a parasite of green cloverworm. Env. Env., 8, 365-368.
- Mason, W. R. M. (1981) The polyphyletic nature of *Apanteles* Foerster (Hymenoptera: Braconidae) a phylogeny and reclassification of Microgastrinae. *Mem. ent. Soc. Canada*, 115. 1—147.
- McCutcheon, G. S. & S. G. Turnipseed (1981)

  Parasites of lepidopterous larvae in insect resistant and susceptible soybeans in South Carolina. Env. Ent., 10, 69—74.
- Moussa, M. A., M. A. Zaher & F. Kotby (1960) Abundance of cotton leaf worm *Prodenia litura* (F.) in relation to host plants. I. Host plants and their effect on biology (Lep. Agrotide Zenobiinae). *Bull. Soc. Ent. Egypte*, 44, 241—251.

- Mueller, A. J. & S. Kunnalaca (1979) Parasites of green cloverworm on soybean in Arkansas. *Env. Ent.*, 8, 376—379.
- Muesebeck, C. F. W. (1921) A revision of the North American species of Ichneumon flies belonging to genus *Apanteles*. U. S. Nat. Mus. Proc., 58, 483—576
- NAGARKATTI, S. & SATYPRAKASH (1974) Rearing of *Heliothis armigera* (Hb.) on artificial diet. *Tech. Bull. Commonw. Inst. Biol. Control.*, 17, 169—173.
- RAO, V. P., M. A. GHANI, T. SANKARAN & K. C. MATHUR (1971) A review of the biological control of insect and other pests in South-east Asia and the pacific region. *Tech. Bull. Commonw. Inst. Biol. Control.*. 6, 46.

# STUDIES ON THE HOST SELECTION AND DISCRIMINATION BEHAVIOUR OF DIAERETIELLA RAPAE (M'INTOSH), A PARASITOID OF LIPAPHIS ERYSIMI (KALT.)

#### S. C. DHIMAN & VINAY KUMAR

Entomological Research Laboratory, Department of Zoology, M. S. College, Saharanpur, India 247 001

(Received 23 September 1985)

The female *D. rapae* walks randomly on the aphid infested plant and discovers its host by macro-orientation. The higher host instars and the adult aphid show stronger defensive reactions in comparison to the younger ones. Contacts of the female parasitoid with aphid were recorded into three categories, 1. mere encounters; 2. encounters followed by ovipositional attempts; 3. encounters followed by actual oviposition. The alate stage of the host is found less parasitized while the half grown nymphal stages are more parasitized.

(Key words: Diaeretiella rapae, host selection, parasitoid, Lipaphis erysimi, oviposition, encounters)

#### INTRODUCTION

Diaeretiella rapae (Hymenoptera: Braconicae: Aphidiidae) has been reported as an internal parasitoid of mustard aphid, Lipaphis ervsimi (Homoptera: Aphididae) from India by Kundu et al. (1965). Later on, the parasitoid has also been reported from this aphid species by ATWAL et al. (1969) and DHIMAN and KUMAR (1983). KUMAR (1985) mentioned that D. rapae causes considerable mortality of mustard aphid and can be used as a bioagent in the control of this aphid. Though considerable amount of work has been done on this parasitoid by various workers, viz., ASKARI et al., 1979; HAFEZ, 1960, 1961; SEDLAG, 1964; TAKADA, 1976. no studies have so far been conducted on the host selection and discrimination behaviour of the parasitoid and present investigations are an endeavour in this direction.

#### MATERIALS AND METHODS

At Saharanpur, D. rapae parasitizes the host L. erysimi on radish plant heavily in comparison to mustard and hence the former plant is selected for culturing the host aphid. The radish plants were cultivated in the earthen pots. The fresh plants were kept in wire gauze cages  $(1 \times 0.5 \times 0.5 \text{m})$  under field conditions during April, 1984. The aphids were cultured on these plants. The parasitoids were reared in glass vial on 30% honey solution separately at 22.5°C. The females of D. rapae were selected and kept together with the males in another separate vial to allow the mating. The male became exited by the presence of female within a minute and he approaches the female by tapping or touching her antennae or other body parts due to which the female became exited and allowed the male to mount on her back. Courtship behaviour lasted 30 to 120 seconds after which the male separated off. After mating, the females were taken out in separate vials using a small aspirator and fine camel hair brush and then transferred into the petridishes having all stages of the host on healthy radish leaf pieces. Transfer of the parasitoids in the petridish was done by slight opening of the lid from one side and making the contact of the mouth of vial with the slit and plugging the rest of the slit with cotton wool. Thereafter, the vial and cotton wool were removed and the lid was closed. The observations on host selection and discrimination were made by using hand lens and under bionocular microscope.

To determine the host instar preference, 2 to 4 hour old mated parasitoid females were transferred on the population of host aphid, L. erysimi, in caged potted plant of radish at 25.2°C and 63.24% R H with photoperiod of 11.00 hours under field conditions. One mated and fully fed female parasitoid was released in each experimental cage. This is slightly advantageous because all stages of the host are present in large number and the influence of laboratory factors are practically excluded. Later on, the mummified aphids were collected from each cage and the percentage of various instars was determined. The data are recorded in Tables 1 and 2.

#### RESULTS AND DISCUSSION

The result of the observations, concerning with the host selection are presented below.

- 1. Host searching—D. rapae drums the radish leaf surace with her antennae while working. It is not yet known whether she detects her host by seeing, smelling or feeding. The parasitoid walks ranon infested leaf surface and domly discovers its host by macro-orientation, ie., the antennae are held forward and slightly bent downwards. If the host, L. erysimi is tapped, the antennae are held upward and the parasitoid wasp may sting the host with her ovipositor and lay eggs. Oviposition takes place than one minute which within less makes difficult to determin what happened actually.
- 2. Host ability, mobility and suitability— Defence reaction and modifications of

the host against the parasitoid attack are rather feeble and they never influence the preference of different host instars by infestation. The higher host instars (IV and V) and the adult aphid show stronger defensive reactions using especially their legs, cauda, antennae and running off.

The female parasitoid prefers the moving aphid, i. e., showing slight movement. It was commonly observed that the female *D. rapae* ignored such aphids that were sitting entirely motionless on plant surface. It is a point of sigificance that the host aphids, *L. erysimi*, remain relatively immobile and however, they show feeble response of female parasitoid when sitting on the plant surface.

Contacts of the female parasitoid with aphids were recorded and divided into three categories:

- 1. Mere encounters.
- 2. Encounters followed by ovipositional attempts.
- 3. Encounters followed by ovipositional attempts and actual oviposition.

There is a difference in the number of encounters in different stages of the host (I to V instars and adults). The parasitoid makes random movements on the substrate, encounter with a host is only a matter of chance. There is also a great difference between various stages of the host encountered by the parasitoid. In the field conditions in most cases where the different stages of the aphid were present together in various colonies and if a colony is encountered by the parasitoid then the individual host stage of this colony will have more chance to be encountered, in place of other stages of the host.

TABLE 1, Preference of Diaeretiella rapae to different stags of Lipaphis erysimi.

	Series-A	-A		series-B	-B		series-C	9		series-D	Q-	
Particulars	Apterous	young	total	apterous	young	total	apterous	young	total	apterous	young total nymph	total
Number of enocunters.	246	144	390	281	261	542	223	170	393	207	205 412	412
Number of encounters with another host with oviposition attempt.	with 136	77	213	110	123	233	100	82	185	101	80	181
Number off eegs deposited.	40	53	93	34	88	119	35	42	77	30	15	45
Number of exposed host parasitoid,	st 27	4	89	24	54	78	24	29	53	23	8	28

TABLE 2. Percentage of encounters, oviposition and egg deposition of Diaeretiella rapae.

	series-A	-A	series—B	_B	series-C	0	series-D	_D
Partiulars	apterous	young	apterous	young	apterous	young	apterous	young
Percentage of total encounters.	63.08	36.92	51.84	48.33	56.76	43.25	50.23	49.73
Percentage of effectiveness of encounters,	16.21	36.80	12.09	32.56	16.59	24.70	14.49	7.32
Percentage of effectiveness of oviposition attempts.	29,41	68.83	30,90	16.69	35.00	49,41	29,70	18.75
Percentage of exposed host parasitoid,	27.00	41.00	24.00	54.00	24.00	29,00	23.00	5.00
Percentage of total egg deposited,	43,01	56.88	28.57	71,43	45.45	54.54	99*99	33,33

Having encountered the host, the parasitoid may either start oviposition or rejection of host may also take place without efforts to oviposit. The latter phenomenon occurred in 47.0—61.0% cases without any difference in various stages of the host. Perhaps, this rejection is due to some intrinsic factors of the parasitoid. The rejection may also take place after ovipositional attempts and it is different in various stages of the host. The effectiveness of encounters can be calculated by the following formula:

Total number of actual oviposition × 100%

= Total number of eggs deposited Total number of encounters × 100%

From this calculation, it is proved that the effect of parasitization is different in various stages of host, i. e., for apterous adult ranged from 14.41 to 16.21% while the alate adults were even less affected 7.32% (Table 2). However, the nymphal instars were more successfully parasitized; the fraction of encounters in oviposition were 36.80% for the first and second nymphal instars, 32.56% for the third and fourth nymphal instars and 24.70% for the fifth nymphal instars. The parasitoid prefers to oviposit in the earlier stages of the host.

During the process of oviposition, wild movement, takes place by the attacked host to scare off the parasitoid. The effect of the movement increased with the development of the host stages. In the more advanced stages, host requires higher number of ovipositional attempt. The effectiveness of ovipositional attempts can be calculated by the following method:

Total number of actual eviposition Total number of oviposition attempts

= Total number of eggs deposited Total number of oviposition attempts

The effectiveness of ovipositional attempts decreased from high value 69.91% for half grown nymphal instar (III to IV) to 29.70 to 35.00% for apterous adult while in the alate adult it is only 18.75%.

The above results support the conclusion, that the frantic movements by the adult host attacked are more effective in scaring off the attacking parasitoid. Another thing is the ability of parasitoid to pierce the integument of the young host is more easy in comparison to hard and thicker integument of the advanced host.

The experiment shows that proceeding from small host to large host stages, the possibility of parasitization increases but the chance of being parasitized decreases. Due to this opposite effect, the half grown nymphal instars are being more parasitized than the other stages of the host. This experiment also indicates that if the hosts are parasitized at random, the process of oviposition and parasitism is not necessary, may be at random. Some stages of the same host species have more parasitism and more parasitoid eggs than some other stages.

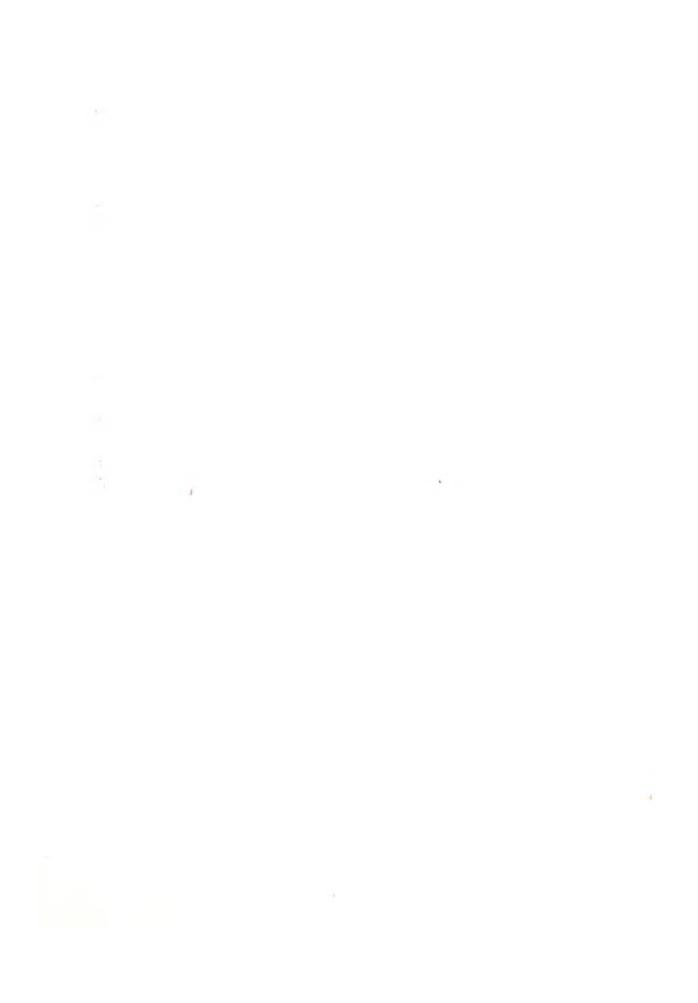
The alate stage is only the stage which has less parasitization, few individuals are parasitized and fewer number of eggs are deposited in this stage. The alate aphids frighten away the parasitoid by wing vibrations and antennal movements or by kicking off or it may fly away to other place.

Acknowledgements: The authors are grateful to Dr. G. E. J. Nixon of Commonwealth Institute of Entomology, London, for the identification of the parasitoid and Dr. G. D. GARG for valuable suggestions.

#### REFERENCES

- Askari, A. & A. Alishah (1979) Courtship behaviour and evidence for a sex pheromone in *Diacretiella rapae* (Hymeneptera: Braconidae), the cabbage aphid primary parasitoid. *Ann. ent. Soc. Amer.*, 72, 749-750.
- ATWAL, A. S., J. P. CHAUDHURY & M. RAMJAN (1969) Some preliminary studies on the bionomics and rate of parasitization of *Diaeretiella rapae* Curtis (Braconidae: Hymenoptera) a parasite of aphids. *J. Res.* (Suppl.), 6, 117—182.
- DHIMAN, S. C. & VINAY KUMAR (1983) Host preference of Aphidius rapae (Curtis) (Hymenoptera: Braconida), Proc. Symp. Ins. Ecol. & Resource Managment, 138—141.
- HAFEZ, M. (1960) Some biotic factors affecting the population of cabbage aphid, *Brivicoryne* brassicae L. in the Netherlands in 1959.

- Meded. 12 intern. Symp. Phytopharmacy and Phytitary Gent., 1347—1350.
- Hafez, M. (1961) Seasonal fluctuations of population density of the cabbage aphid, Brivicorynae brassicae (L.) in the Netherlands and role of its parasite Aphidius (Diaeretiella) rapae (Curtis). T. Pl. Ziekter, 67, 445-548.
- Kumar Vinay (1985) External morphology and ecology of *Aphidius rapae* (Curtis) together with studies on biological control of its host, *Lipaphis erysimi* (Kalt.). Ph.D. Thesis submitted to Meerut University, Meerut.
- Kundu, G., G, V. K. SHARMA R. K. ANAND & S. RAI (1965) New record of *Diaeretiella* rapae (Curtis) as a parasite of mustard aphid, Lipaphis erysimi. Indian J. Ent., 27, 497-498.
- SEDLAG, S. (1964) Zur Biologie und Bedeutung von Diaeretiella rapae (M'Intosh) als parasit der Kohlblattlaus (Brevicoryne brassicae L.). Nachrbl. Deut. Pflanzen. Schutzdienst., (Berlin) 18, 81-86.
- TAKADA, H. (1976) Studies on aphids and their parasites on cruiferous crop and potatoes. I. Parasite complex of aphids. Kontyu, 44, 243—253.



### IN MEMORIUM

P. P. GRASSE (1895-1985)



Professor Pierre-Paul Grasse whose name is famous throughout the scientific world as the editor of the 37-volume Traite de Zoologie (a real encyclopaedia of Zoology, each volume comprising from 800—1000 pages or more, giving the most modern synthesis of our knowledge on the various animal groups from Protozoa to Primates), the founder of the international journal' Insectes Sociaux', the most eminent termitologist the world has produced, and who left his indelible impression on the diverse areas of biology he touched. breathed his last on 9th July 1985 at the age of ninety years,

Grasse was drawn indirectly to the study of termites through his earlier research (1924 onwards) on the flagellate Protozoa of the lower termites and the nature of this termite-flagellate symbiosis with special reference to its implications for their termite hosts. From then onwards the termites were for a period of half a century his most favourite area for research. This enabled him to bring out a 3-volume treatise 'Termitologia' (2044 pages), of which the first volume appeared in 1982, the second in 1984 and the third, posthumously in 1986. There are no aspects in the biology of the termites to which he had not made important contributions. He had often stated that nothing can equal the close observations made on the animal in the natural milieu. This made him undertake several expeditions mostly to tropical Africa where the termite fauna is very rich in species as well as in their behavioural diversity.

Grasse clearly demonstrated the importance of the proctodeal exchange of food (totally different from excrements) among the worker castes or workers and older larvae and its social implication as an important factor contributing to the maintenance of colony life. Digging into the very large cathedral-shaped mounds of Macrotermes natalensis in the African Savanas, Grasse was impressed by the great volume of the fungus gardens and of fungus combs each contained. His later studies established the fact that the fungus combs along with the specific exclusively termitophilous fungus (Termitomyces) formed a regular part of the food of the fungus-cultivating higher termites Grasse and his stu-(Macrotermitinae). dent and close associate Noirot (1959) proved that the enzymes present in the fungi served to predigest the lignin of the fungus combs into easily digestible They also demonstrated that products. while the lower termites show symbiosis with their rectal flagellates, the funguscultivating higher termites show a double symbiosis, firstly with the bacteria contained in their gut and secondly with the specific fungus Termitomyces, both enabling these termites digest their cellulosic diet. In 1978 at the age of 83 years (!), Grasse was the first to conclusively show the real nature of the fungus combs by demonstrating the existence of two types of transits through the termite intestine: one which takes place at a rapid pace, starting with the raw food and giving rise to the material with which the combs are made; the other at a slow pace, starting with the comb material (already partly digested by the fungi) which underwent normal digestion. Grasse and Noirot (1948) were also the first to report the process of formation of new colonies by the division of the existing colony into several groups and the development of each group into separate viable colonies, a process they termed 'Sociotomy'

Termites, according to Grasse, are indefatigable movers of soil. The construction of epigeous mounds necessitates the bringing up of a considerable mass of soil from various levels underground. He has also been the first to point out (1950) the importance of termites in the origin and development of tropical soils.

The origin of polymorphism and caste differentiation in termites have always been problems to which Grasse gave much thought and he realised the great significance of these phenomena in the social life of termites as well as of the other social insects. The termites show great diversity in these processes and it was Grasse who reported the existence of 'achrestogonimes' and of the 'pseudergates' He also pointed or the false workers. out peculiarity of the soldier caste by showing their total dependence on the society (the workers or the older larvae) for feeding them, for the soldier is incapable of feeding by itself (1939). The study of the origin of polymorphism and caste differentiation in the higher termites (Termitidae) which defied all earlier efforts to yield satisfactory explanation, was successfully worked out by Noirot (1955) under Grasse's supervision.

Deeply convinced about the unity of the living world in spite of its extraordinary diversity, Grassé was the first to point out the essential common characteristics of all animal societies while at the same time giving due importance to their polyphyletic nature. He recognised that an interattraction of an olfactory nature among individuals of a colony forms the most important principle of which all forms of social life is maintained. He also showed that social life would result in the emergence of new potentialities for the colony by its consequences on the individuals forming the colony as a result of what he called 'effet de groupe' (group effect) (1946). In this phenomenon of group effect, Grasse asserted that the actions of congeners in a group act through specific sensory channels to modify the behaviour, physiology or even the course of development of individuals. Such complex interactions established among the members of the society bring about a true homeostasis through the action of social regulations (1949). For this the termites furnished him with the most appropriate examples, as such social regulations are operative particularly during the formation of the replacement reproductives as a result of the removal of the functional reproductives.

Grasse wondered how the blind worker termites possessing only limited havioural repertoires and in which the isolated individual by itself is incapable of even continued existence, become capable when in a group, of constructions at times of very gigantic proportions, such as the mounds of M. natalenis, possessing a definite plan, a geometric shape and a specific architecture. Rejecting all metaphysical interpretations, for a long time he searched for a 'directing influence' and at last in 1959 arrived at the concept of 'stigmergy'. This he deduced from his earlier observations on the reconstruction in M. natalensis of the royal cell, with the queen in situ. He found that the workers began to deposit pellets of earth haphazardly at a certain distance from Where the pellets of earth the queen. happened to form heaps, these now developed new qualities and meanings and acted as the foci for building behaviour, resulting in the construction of The workers finally connected the adjacent pillars with each other in such a way that it formed a continuous wall. It is thus seen that to begin with, the co-ordination of individual activities is indirect. It is the product of the work already completed which incites and directs further work until the whole species-specific product of the construction behaviour emerges. Grasse's studies (1981) on polycalic nests of Apicotermes lamani and of its construction behaviour led him to conclude that all collective functions such as nest construction or its repair and the regulation of such activities even in cases which necessitate a close co-ordination of individual acts and which may even show semblances of anticipated behaviour, are governed by very clear 'automatism' (power of initiating vital processes originating at the cellular level) which he designated as instinctive complexes'.

Grasse never liked to be limited to just one or two specialisations in research. His insatiable curiosity urged him to take up research in diverse areas of the entire field of contemporary zoology & general biology including evolutionary biology and his nearly 400 publications bear ample testimony of his great versatility. He was an extraordinary promoter of new ideas and of new lines of research and used to make his students further pursue the new lines of research he had initiated. A protozoologist and cell biologist of eminence, Grasse was highly competent in the study of the ultra-structure of cell organelles, so much so, during the 1950s, an epoch when the use of the transmission electron microscope made possible the study of cell organelles and structures at ultra-high magnification, the National Centre for Scientific Research (CNRS), Paris, gifted him with an electron microscope laboratory which formed the nucleus for ultra-structural studies by a distinguished group of researchers under his direction

The several expeditions which took Grasee to the heart af tropical Africa for the study of the biology of termites, also enabled him to develop a passion for primatology. There in Gabon he founded in 1967 (?) the 'Laboratory of Tropical Biology' where groups of research workers carry out studies on the psychology and behaviour of the anthropoid apes and on the behaviour of various other animal groups.

But the discipline which Grasse most preferred and in which his contribution was most prolific was that of ethology and the psycho-physiology of the social insects, for which he suceeeded in forming a strong French school of researchers. At the Internationan Congress of Entomology, Amsterdam (1951) he founded the 'International Union for the Study of Social Insects' and he and Professor Gösswald (Würzburg, FRG) took the initiative for forming a separate secion for Social Insects at international congresses. In 1954 he again was the prime moving force for starting prestigious journal 'Insectes Sociaux' which has by now published 33 volumes.

On the occassion of Prof. Grasse's retirement on 30th September 1967, his friends, colleagues, admirers and past an present studeuts cerebrated his 50 years of very distinguished service in the cause of science, under the presidentship of the Minister of State in charge of scientific researh. There he

declared that he shall continue his research, his writing and editing work and that his mind is full of new projects but of which he is not sure of accomplishing all. In January 1982 at the age of 87 years he wrote to me (his doctoral student of 1954—58): "I shall continue to work till there is life left in me".

It was but natural that Grasse's fame spread far and wide. He became the unnamed ambassador abroad of the french scientific and teaching community. He was the highly respected President of prestigious Academy of Paris, the Honorary President of the 'Societé Entomologique de France', An Associate Member of the Royal Academy of Belgium, Vice-President of the International Congress of Zoology and Honorary Member of the International Congress of Entomology. The Universities of Brussels and of Gand awarded him the degree of Doctor of Science honoris causa. Wherever he went outside France, his fame and reputation preceded him. He was invited to visit most of the european countries where his learned audiences benefited from his inexhaustible fund of knowledge. In Brazil, the scientific community received him most enthusiastically and his lectures elicited such great admiration and respect that several Brazilian collaborators accompanied him to his laboratory in Paris to work under his enlightened supervision. The International Congresses of Zoology and of Entomology held in the U.S.S.R., the U. S. A., Denmark, Germany, England, Canada and elsewhere listened to his talks with great interest and admiration.

Pierre-Paul Grass'e was a very enthusiastic professor, dynamic speaker, a born naturalist who loved equatorial Africa, researcher of eminence, methodical and perseverent observer, alert writer and editor, severe critic and a true patriot of his great country and an exponent of its great cultural heritage. He shall be remembered as one of the greatest biologists France has produced,

and without exaggeration, as one of the greatest zoologists of all time. May his noble example of devotion to scientific research and the dedication of his entire life-time to the advancement of biology serve to inspire us all.

K. J. JOSEPH

Professor & Head of the Department of Zoology, University of Calicut, Calicut University P. O., Kerala, India 673 635

#### **BOOK REVIEW**

INSECTS AND MITES OF CROPS IN INDIA. by M. R. G. K. NAIR, Publication and Information Division, Indian Council of Agricultural Research, New Delhi, 1986, 408 pp., Figs. 301, Price Rs. 42.50.

This is the revised edition of the book published earlier in 1975. In this edition is included a considerable amount of information on the insect and mite pests of crops in India and their control which has accumulated since the date of its first publication. A new chapter on pests of fodder crops and a new section on pests of Cacao also have been added. A list of scientific names arranged alphabetically is a new feature of this edition. With the revision, the value of this book as a reference book on crop entomology and acarology in India has increased.

A. VISALAKSHY

## Statement of ownership and other particulars about ENTOMON

(Form IV, Rule 8 of the Registration of Newspapers (Central) Rules, 1956)

1. Place of publication: Trivandrum

2. Periodicity of publication: Quarterly

3. Printer's name, nationality and addrers : V. K. Kesava Prabhu, Indian,

Department of Zoology, University of Kerala, Kariavattom, Tvm - 695 581.

4. Publisher's name, nationality and address: —Do—

5. Editor's name, nationality and address : —Do—

6. Name and address of individual who

owns the newspaper

: Association for Advancement of Entomology, Department of Zoology, University of Kerala, Kariavattom,

Trivandrum - 695 581.

I, V. K. Kesava Prabhu, hereby declare that the particulars given above are true to the best of my knowledge and belief.

(Sd./)

Trivandrum, March 31, 1987. Dr. V. K. KESAVA PRABHU Publisher, Entomon. bove

#### **ANNOUNCEMENTS**

### IV Oriental Entomology Symposium

The IV Oriental Entomology Symposium is being planned to be held in November 1988 in Agra. There will be scientific sessions and workshops on specific topics. Scientific sessions will have the following major sections: insect systematics, morphology and ultrastructure, biology and ecology, physiology, medical and veterinary entomology. It is also planned to have workshops on specific themes and topics like insect behaviour, social insects, insect control, high altitude biology, Agromyzidae, Chironomidae, parasitic Hymenoptera, host specificity and host selection etc. Facilities will be provided for organising workshops on specific themes of importance. Those who are interested in participating should contact Dr. Ipe M. Ipe, Convener, IV Oriental Entomology Symposium, School of Entomology, St. John's College, Agra.

IPE M. IPE

### V All India Symposium of Invertebrate Reproduction

V All India symposium of Invertebrate Reproduction will be held at P. G. and Research Department of Zoology, Arulmigu Palaniandavar College of Arts & Culture, Palani, Tamil Nadu during the 4th week of September, 1987. It is being organized by the Indian Society Invertebrate Reproduction. Contributions in any area of invertebrate reproduction are welcome. One of the focal themes proposed in the symposium is the productive biology of invertebrates (Aquaculture, Sericulture, Apiculture etc).

For further details write to:

Prof. Dr. S. PALANICHAMY, Convener, V All India Symposium of Invertebrate Reproduction, P. G. & Research Department of Zoology, Arulmigu Palaniandavar College of Arts & Culture, PALANI, Tamil Nadu - 624 602 ENTOMON is covered in the following abstracting/indexing journals: Chemical Abstracts (Chemical Abstracts Service, The Ohio State University, Columbus, Ohio 43210, U.S.A.), Review of Applied Entomology (Commonwealth Institute of Entomology, 56 Queen's Gate, London SW9 5JR, England), Science Citation Index and Current Contents Agriculture, Biology & Environmental Sciences (Institute of Scientific Information, 3501 Market Street, Philadelphia, Pa. 19103, U.S.A.), Biological Abstracts (Biosciences Information Service, 2100 Arch street Philadelphia, Pa. 19103, U.S.A.), Entomology Abstracts and other relevant Abstracts (Information Retrieval Limited, 1 Falconberg Court, Lodon WIV 5FG, England), Referativnyi Zhurnal (The Institute of Scientific Information, Academy of Science of the U. S. S. R., Baltijskaya ul., 14, Moscow A-219, U. S. S. R.), Current Advance in Biological Sciences, 132 New Walk, Leicester LEI 7QQ, England.